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BULLETIN

OF THE

Harvard University.

MUSEUM OF COMPARATIVE ZOÖLOGY

AT

HARVARD COLLEGE, IN CAMBRIDGE

VOL. LIII.

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AT HARVARD COLLEGE.
Vol. LIII. No. 1.

THE LIGHT RECIPIENT ORGAN OF THE COPEPOD, EUCALANUS ELONGATUS.

By Calvin Olin Esterly.

With Six Plates.

CAMBRIDGE, MASS., U. S. A.:
PRINTED FOR THE MUSEUM.
October, 1908.

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No. 1. — CONTRIBUTIONS FROM THE ZOÖLOGICAL LABORATORY
OF THE MUSEUM OF COMPARATIVE ZOÖLOGY AT HARVARD
COLLEGE, UNDER THE DIRECTION OF E. L. MARK, No. 196.

The Light Recipient Organs of the Copepod, Eucalanus elongatus.

By Calvin Olin Esterly.

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I. Introduction and Methods.

Although the decapod Crustacea have been the objects of much careful morphological investigation and experimentation, in the fields of comparative neurology and psychology, the organization of the nervous system of the lower Crustacea has been almost entirely neglected in the neurological studies which have been made in the last fifteen years.

Since the appearance of the papers by Richard (**'91**) and Claus (**'91**), practically no study has been made of either the central or peripheral nervous system of the Copepoda, although the literature which deals with the group mainly from a systematic standpoint may contain scattered statements concerning the nervous system and sense organs. Even the little work that has been done deals almost exclusively with the gross anatomy of the nervous system. Such matters as the fibre tracts and systems of neurons have received the most fragmentary and superficial treatment. Work of this kind must now be regarded as relatively unsatisfactory because of the advance in general technique and neurological methods during the last few years. A thorough, painstaking study of any one of the Copepoda is worth while because of the comparatively low organization of the group, both for the sake of comparison with the higher forms of Crustacea and for a knowledge of the group itself.

Within the group of Copepoda the marine forms have been least studied, for the paper of Richard ('**91**), which is the most thorough of any dealing with the more minute anatomy of the nervous system, is concerned with only fresh-water forms. For twenty-eight years previous to the appearance of Richard's work there was none dealing primarily and specifically with the nervous system. Claus ('**63**) devoted a portion of his monograph on the non-parasitic Copepoda to the anatomy of the marine representatives, and in it we have the most complete, as well as the best, comparative study of the nervous system and sense organs of the pelagic forms that has appeared.

The general disregard of the nervous system in the marine Copepoda would alone be reason enough for taking up the study at the present time. But in view of the closer attention which the nervous apparatus in decapods has received recently, a study of the lower forms for purposes of comparison seems especially desirable. The present investigation grew out of a desire to gain some knowledge of the peripheral nervous system and sense organs of the Copepoda as a preliminary step toward a study of the reactions, including the diurnal and seasonal movements, of these enormously abundant and very important plankton organisms. It is hoped that this paper may be one of several dealing with the nervous system, the central organs being taken up at a subsequent time.

For this purpose one of the very commonest calanoid copepods found in the waters of the Pacific Coast was selected as a basis for study. This form is Eucalanus elongatus Dana. It is particularly suitable for work in which microscopical methods are to be employed, because it is of large size, and exceedingly transparent; moreover the chitin is so thin and delicate that it does not cause the least trouble in cutting paraffine sections. The treatment ordinarily given material intended for microscopic investigation is sufficiently satisfactory in this case, but to prevent shrinkage and distortion, care is necessary in the use of the higher grades of alcohol, in clearing and in transferring to paraffine.

In dehydrating, I have made it a practice to transfer the material from 90% alcohol to 95% alcohol and then after 5 or 10 minutes to replace this by absolute alcohol. Xylol has proved to be the best clearing agent. If this is added drop by drop to the absolute alcohol until the mixture has reached the proportions of $\frac{1}{3}$ alcohol and $\frac{2}{3}$ xylol, and then pure xylol is used, the animals will clear quickly and without the least shrinkage. Before transferring to the paraffine used for imbedding it is well to allow the object to remain for a time in a saturated solution of paraffine in xylol. I have always used for imbedding purposes a paraffine which melted at 60° C. and sections were generally cut 10 μ in thickness, though sections 6 μ in thickness are readily obtained. It is not necessary to employ a reagent for softening or destroying the chitinous covering of the animal, for, after any fixation employed, perfect sections and ribbons can be obtained. The chief trouble has been to preserve the animal in its normal form, so that it might be oriented for cutting sections in particular planes. Dehydration and clearing take place very rapidly, but infiltration with paraffine is slower. It is well to leave the animals in melted paraffine for an hour or a little longer.

I have relied chiefly upon a modification of one of vom Rath's mixtures and upon Zenker's fluid for killing and fixing reagents. Saturated aqueous corrosive sublimate and a corrosive-acetic mixture have likewise been employed, as has also 10% formalin; but Zenker's fluid is by far the most satisfactory of all these reagents. The osmic acid mixture of vom Rath was modified slightly in the amount of platinic chloride. To a mixture containing 12 c. c. of 2 % osmic acid, 100 c. c. saturated aqueous solution of picric acid, and 1 c. c. of acetic acid, I added 25 c. c. of a 0.2% solution of platinic chloride. This has given satisfactory results. It has been allowed to act for varying lengths of time upon animals put into it from sea-water, and has in some cases been followed by pyroligneous acid, as recommended by vom Rath ('**95**). I believe it is better to allow the fixative to act alone and for not more than 36 hours, otherwise the blackening is so great as to detract from the value of the preparation. When the fluid is used for that time without further treatment the medullated nerve fibres are blackened intensely, while the rest of the nervous system is a deep brown. If pyroligneous acid is used subsequently to the platinic-chloride mixture, all parts of the animal become so intensely black as to be useless, and the tissue is rendered extremely brittle as well. The material may, however, be very readily decolorized to any extent by the use of peroxide of hydrogen. Very good preparations may be obtained in this way, either from tissue which is too dark from the action of the picric-osmic mixture alone or from that which has in addition been treated with pyroligneous acid; the method may be applied to the entire animal, or to sections upon the slide. Decolorization may be controlled by watching the tissue during the process. I have used the pure commercial peroxide without visible injury to the tissue, but the rapidity of decolorization depends upon the strength of the solution employed.

The stains I have used have been chiefly the iron-haematoxylin of Heidenhain, Mallory's (:00) connective-tissue stain, and the platinic-osmic-acetic mixture of vom Rath, which was also the fixing reagent. The three stains are about equally valuable; each supplements the others. The haematoxylin is best for nuclear structures; vom Rath's fluid is excellent for nerve-tracts and medullated fibres. The connective-tissue stain of Mallory is unexcelled for general contrast and for extreme delicacy in many structures, such as the nerve-endings in the cells of the eye. It is very serviceable in any tissue unless the nuclei are to be particularly studied. It differentiates nerve tracts in the brain as well as, or better than, vom Rath, since the color contrast is greater, but for following the course of single fibres it is in general not as satisfactory as the latter. Mallory's stain has the disadvantage, possibly, that it is rather capricious; at any rate preparations in which it has been used vary greatly in value and unaccountably so. But when successful, the stain gives pictures which are most excellent in the matters of delicacy and sharpness. In addition to this there is the great advantage of simplicity and rapidity of procedure in its use.

The methylen-blue method has, in my hands, been uniformly unsuccessful in the case of the marine copepods, though it has been tried many times. It is, of course, impossible to inject the stain, and if the animals are immersed in a solution the result has been negative. The marine forms do not respond to such treatment in the way that the fresh-water forms do (Esterly :06).

No attempt has been made to impregnate nervous structures according to the method of Golgi, and I have not obtained good results from the Bielschowsky method.

Another method of preparation which has given excellent results and, I believe, offers unique opportunities for study, is that of mounting the central nervous system and sense organs entire. This is made possible by the fact that Eucalanus reaches a length of from 6 to 8 mm. The entire dorsal portion of the body may be removed by a stroke of the scalpel, thus leaving the nervous system, including the eye and a large part of the nerves, in place upon the ventral body wall. Such preparations may be cleared and mounted in the usual ways, and if enough of the body has been removed, the preparation will be of such slight thickness that high powers of the oil-immersion may be used without injuring the specimen. If the staining, or coloration, due to vom Rath's mixture is not too deep, one may follow single nerve fibres for long distances, both within the cord and outside it; and in any case the ganglia and nerves in the thoracic segments stand out with almost diagrammatic clearness. In the region of the mouth the muscles of the organs pass toward the median plane of the body and overlie the cord so closely that it is impossible to remove them without injury to the nervous structures. This disadvantage may be overcome to a considerable extent by comparing many entire preparations and supplementing them by series of frontal sections.

Before passing to a description of the histological features of the light recipient organs in Eucalanus, I wish to express my appreciation of the guidance and criticism of Prof. E. L. Mark, under whose direction the work has been done. His continual attention to accuracy of observation and deduction, and his conservative judgment, have been invaluable. I also wish to acknowledge the opportunities enjoyed at the laboratory of the Marine Biological Laboratory of San Diego, where all of the material was obtained, and where, except for the staining and sectioning, it was all prepared.

II. The Optic Apparatus.

1. The Median, Uninverted, Eye.

The eye of Eucalanus belongs to the type which has been designated by many authors as an "x-shaped pigment spot". Excepting the investigations of Richard (**'91**), Claus (**'91**), and Hartog (**'88**), there appear to have been none based upon a study of microscopic sections of the tripartite eyes in the Copepoda, unless we except the work of Hesse (**:01**), which deals particularly with the type of nerve ending in the cells rather than with the structure of the eye as a whole. Most investigators have based their studies upon the fresh-water forms, such as Cyclops or Diaptomus, while but three, so far as I am aware, have had an opportunity to study the marine forms.

Claus ('**63**) mentions no features of the eye of Eucalanus (E. attenuatus Dana) except its general form, the optic nerve, and the relation of the frontal nerves to the eye. Grenacher ('**79**), working upon the same form, made a closer study, but employed only isolation preparations or entire ones. His observations were confirmed in some respects by Claus ('**91**). Hesse (:**01**) was the first to make sections of the eye of Eucalanus.

Grenacher ('**79**, p. 63) has given an accurate account of the optic organ. He found that it conformed to the well-known tripartite type, or "x-shaped pigment spot," consisting of a ventral unpaired portion and two dorsal paired parts, each of the three divisions possessing a special pigment plate. To each of the pigment plates belongs a very constant number of cells, all of which, though not uniform in shape, turn an attenuated end toward the point of exit of the optic nerve, which lies between the three divisions of the eye. Each of the paired portions of the eye, or the "lateral eyes," contains eight cells arranged in two layers. The upper layer contains five cells and the lower layer three. The median, ventral or unpaired eye contains ten cells, eight of which are arranged in pairs, two being unpaired. The optic nerve fibres continue into the cells, entering them at their pointed ends. Grenacher states that the fibres could be traced within the cells in some cases. Claus ('**91**, p. 229) states that Grenacher failed to notice that the nerve fibres enter from the outer side, thus giving the eye the nature of an inverted "Becherauge," but (p. 246) confirms Grenacher's observations as to the number and position of the cells. Hesse (:**01**, p. 350) mentions the eye of Eucalanus very briefly, merely pointing out the character of the nerve endings and the presence of the "interior bodies" (Binnenkörper).

I have studied the eye of Eucalanus both by means of sections and from preparations of the entire organ. The general form of the eye is shown in Plate **1**, Figure 1, and has already been described by Claus ('**63**, Taf. 7, Fig. 9) and by Grenacher ('**79**, Taf. 5, Fig. 36; Taf. 6, Fig. 37, 38) in E. attenuatus. I have been unable to obtain any specimens of this species, but, so far as I can judge from these descriptions, the shape and more general features of the eye are the same as in E. elongatus.

a. *Location and General Features.*— The organ is on the average about 440 μ in front of the anterior end of the brain. It lies in the sagittal plane of the body and the ventral component rests upon the chitin of the ventral wall of the body, except for the intervention of an extremely delicate enveloping membrane (Plate **5**, Fig. 44).

It will be seen that the lateral paired eyes (Fig. 1) are oval and cup-like, the longer axis lying in the direction of the main axis of the animal's body. The lateral margins of the paired eyes extend beyond the lateral borders of the median ventral eye (Fig. 1, *oc. m.*), thus obscuring the latter when the organ is viewed as a whole from the dorsal side. When looked at in this direction, the ventral eye may, however, be seen between the paired eyes. The anterior and posterior borders of the three divisions of the eye lie in nearly the same transverse plane of the body.

The cup of the ventral eye is about three-fourths as deep as those of the lateral eyes, but it is about five-fourths as broad in the transverse plane of the body. The whole eye is slightly flattened in the dorso-ventral direction (Fig. 7).

The optic nerve (*n. opt.*) emerges from the eye directly behind, and dorsal to, the ventral ocellus (Plate **1**, Figs. 1, 2, 3, 6; Plate **5**, Fig. 46), between it and the postero-ventral curvature of the lateral ocelli. The nerves from the rostral organ (Figs. 1, 7, *n. f.*)— the frontal nerves of Claus and others — are conspicuous strands, which meet the optic nerve a short distance behind the eye, having passed, from their distribution in the rostrum, over the outer and dorsal surface of the lateral eyes. The rostral nerves do not innervate any of the cells of the eye.

b. *Pigment Mass.*— The central mass of the eye, between the optic cups, consists, I believe, of a single cell (Plate **1**, Fig. 7; Plate **2**, Fig. 23; Plate **5**, Fig. 49, *cl. c.*). I have never seen more than a single nucleus (Plate **1**, Fig. 2, *nl.″*) in this region whatever the method of preparation. Consequently it seems to me that the three optic cups may be said to rest in or upon a central cell (Plate **1**, Figs. 7, 9; Plate **2**, Fig. 23), which, as seen in cross-sections of the eye, is in general triangular. That is, the region between the median walls of the lateral eyes and the dorsal surface of the ventral eye is three sided, and is occupied by a single cell.

In Cyclops, Richard ('**91**, p. 207, Pl. 7, Fig. 23) found that the central pigment mass is composed of three cells separated from one another by two membranes, and Hartog ('**88**, p. 33) speaks of the division of the central mass by fine partitions separating the "blocks" which receive the optic cups; the blocks contain nuclei, at least one, probably two. One would be led to expect similar conditions in the eye of Eucalanus, since it resembles so closely in general features the eye of Cyclops. But in the many series of sections studied, which have been cut in the frontal, sagittal and transverse planes, I have never seen traces of more than one nucleus which could by any possibility be related to the central mass of the eye,— that is, to that portion of the organ which lies entirely outside the true optic cups, and between the three. Moreover, there is no trace of what could be called cell walls or membranes in this region, and it seems unlikely that such would fail to appear in sections that show the boundaries of the retinal cells very clearly.

The conclusion seems justified, therefore, that the median portions of the three optic cups in the eye of Eucalanus are embedded in a single cell, which corresponds to the "blocks" of Hartog (**'88**, p. 33) and to the "masse central de pigment" described by Richard (**'91**, p. 207) in the fresh-water Copepoda.

That this cell contains the pigment of the eye, seems highly probable for a number of reasons, though this location is very different from that assigned to the pigment by Grenacher (**'79**, p. 64). The central cell of the eye (corresponding to Richard's "central pigment mass") is in precisely the same position with reference to the rest of the eye as is the pigment mass in Cyclops. Moreover, its lateral and ventral margins form what Hartog (**'88**, p. 33) has named the "tapetum," which he says "consists of fine reddish granules, lying on the face of the block." I can confirm the portion of the statement relating to the position of what seems to be a reflecting layer in the pigmented portion of the eye of Cyclops, but I have not been able to satisfy myself that the tapetum consists of granules. It seems, rather, to be a differentiated margin of the pigment cell or cells. At any rate, the central cell of Eucalanus elongatus has when sectioned precisely the same appearance as in Cyclops and I believe that the structures are homologous in the two cases. (Plate **1**, Figs. 2, 3 *tap.*; Plate **2**, Fig. 16; Plate **5**, Fig. 49.)

In my preparations the tapetal layer is colored yellowish in Mallory's connective-tissue stain (Plate **5**, Fig. 49), and intensely black in vom Rath's (Plate **1**, Fig. 5). In haematoxylin stains the tapetum has a vitreous appearance both in Clyclops and in Eucalanus, and it is this fact, as much as its position, that has led me to conclude that we are dealing with the same structure in the two forms, and that the single central cell in Eucalanus is the pigment cell of the eye. Such indirect evidence is all that is available, for the actual pigment is not demonstrable in any of my material, probably because of its solubility in alcohol. Parker ('**91**, p. 78) has found that in Pontella the pigment dissolves very readily in alcohol, so that all traces of it disappear under this treatment. I can state positively that the eye of Eucalanus elongatus does contain reddish pigment while the animal is alive, for I have seen it many times, but without noticing closely its location. Giesbrecht ('**92**, Taf. 3, Fig. 1) has also seen such a condition and has figured it. In view of this comparative evidence, I think it likely that Grenacher has mistaken certain other structures for pigment plates, though it is reasonable to suppose from the general appearance of the objects which he called pigment plates, that they, too, have this function. It should be added here that the cytoplasm of the central cell (Plate **1**, Fig. 6, *cl. c.*) is of a vacuolated, spongy appearance, while the plates which Grenacher has described are very dense and homogeneous in appearance, showing no traces of granulation.

c. *Basal Plates.*— Plates like those described by Grenacher are very conspicuous objects in the eye of Eucalanus elongatus (Plate **1**, Figs. 1, 2, 6, 9; Plate **5**, Figs. 49, 50, *la. ba.*), though there are differences between the conditions in this species and those described by Grenacher in E. attenuatus. He states ('**79**, p. 64) that each of the three portions of the eye has "a special pigment plate." In Eucalanus elongatus, each lateral eye possesses two plates, an anterior and a posterior, which, as I have said, do not contain pigment, if such evidence as has been brought forward is valid. The plates of the lateral ocellus cover the median ventral surface of the ocellus. Each plate is triangular in outline, and conforms to the curvature of the cup; in the natural position of the animal the apex of each triangle is invariably directed ventrally (Plate **2**, Figs. 21, 22). The plates are of about equal size, and in general the dorsal margins of the two together extend around the median half of the eye (Plate **1**, Figs. 1, 2). The anterior and posterior plates approach each other, but are never in contact, so far as I have seen. The point where they most nearly touch lies approximately in the mid-transverse plane of the eye.

As seen in cross sections of the eye, each plate occupies what may be termed the ventro-median third of each lateral eye (Plate **1**, Fig. 9). The extent of the plate in such a section is practically the same as the tapetum on that face of the central cell (Plate **2**, Fig. 16; Plate **5**, Figs. 49, 50). In frontal sections (Plate **1**, Fig. 2; *la. ba.* and *tap.*) it may be seen that in an antero-posterior direction also the tapetum is practically co-extensive with the plates.

The plate of the ventral eye (Plate **1**, Fig. 6, Plate **5**, Figs. 49, 50, *la. ba.*) rests as a cap on the dorsal surface of the ventral cup. It may be regarded as a single structure, though it is perforated by openings, through which the nerves pass from the retinal cells (Plate 5, fig. 50; *la. ba.* and *fbr.*). The plate is oval in outline, but does not extend to either the anterior or posterior border of the ventral eye (Figs. 3, 6), nor does it quite reach the lateral margins of that portion of the eye (Fig. 7). The plate of the ventral eye is in all respects similar to those of the lateral eyes, except for the openings in it already mentioned. The description of the latter may be deferred until the discussion of the innervation of the eye is taken up.

It should be added that, although in entire preparations the basal plates of the eyes seem to be very thin shells, they are in reality of considerable thickness. In Figure 49 (Plate 5) the plate as shown in the right lateral ocellus (*ocl. dx.*) has been cut almost perpendicularly to its two broad surfaces. In other figures where the basal plates appear the plane of the section is such that a true idea of the real thickness of the plate is not given. There are two reasons for this: first, it is very difficult in embedding the animals to orient them so that the plane of sectioning will be more than approximately that which is desired; and, secondly, on account of the curvature of the plates, especially those of the lateral eyes, very few if any of the sections coincide with the radius of curvature. In Plate 5, Figure 45, the basal plate of the ventral eye is cut almost perpendicularly, and I believe that this gives a fairly accurate idea of the real thickness of this plate.

We are justified in summarizing the knowledge of the tripartite eye of Eucalanus thus far gained as follows. The paired latero-dorsal and the single median-ventral optic cups rest upon, or in, a single cell, which forms the pigmented background of the retinal elements. In this respect the eye of Eucalanus differs from that of Cyclops or Diaptomus, as described by Hartog ('88) and Richard ('91), where at least more than a single cell exists in the central mass of the eye. In Eucalanus, as in the fresh-water forms, the faces of the central mass (or cell) which are turned toward the optic cups form the tapetal layer; these are to be looked upon as products of the central cell and not as independent cells. The so-called pigment-plates, which Grenacher ('79) described in the eye of Eucalanus, are probably not such, if we may rely upon comparative evidence adduced from the conditions seen in other Copepoda. But whether Grenacher's interpretation is the correct one or not is of less real importance than the fact that pigment is known to occur in the eye. Its location need not necessarily be known for a fair understanding of the morphology of the eye. It seems to me, however, that indirect evidence points toward the central cell as containing the pigmented background of the eye, rather than to the plates on the inner faces of the optic cups, which partially envelope the retinal cells.

d. *Number and Arrangement of the Retinal Cells.*— The optic cups themselves (Plate **1**, Figs. 1, 2, etc.) are more or less globular masses of retinal cells, as Hartog ('**88**), Richard ('**91**) and Grenacher ('**79**) have shown. The last named author determined accurately, as already stated, the number of cells in each part of the eye of Eucalanus attenuatus (Calanella mediterranea). In Eucalanus elongatus the cells in the ventral eye (Plate **1**, Figs. 8, 10; Plate **2**, Figs. 21, 22) are arranged on precisely the same plan as Grenacher ('**79**, p. 65) has described for the other species. That is, there are in all ten cells; of these, one is in almost the exact centre of the eye, lying slightly anterior to the mid-transverse plane (Plate **1**, Fig. 10). There is one cell directly anterior to it, and, on each side of the longitudinal axis of the eye are four others, which are paired, each meeting its mate in the median plane. There are, then, two unpaired and four pairs of cells in the ventral eye. This number can be very readily determined either in frontal sections, where the whole number may be seen in a single favorable section (Plate **1**, Fig. 8), or by reconstructions from sagittal or transverse series of sections. It is not so easy to count the nuclei in entire preparations, but when that is possible there is no doubt as to the number. I have very many preparations of the whole eye, but with a few exceptions the conditions are not favorable for counting the cells in the ventral eye. Figure 21 (Plate **2**) is drawn from a vom Rath preparation which had been decolorized in H_2O_2. The preparation is viewed from the ventral surface, consequently the ventral eye is uppermost in the drawing. It was impossible to see the most anterior nucleus of the ten in the ventral eye, yet the cell walls were perfectly distinct. Likewise the posterior nucleus of the four on the right side (left in the drawing) was invisible, yet from the arrangement of the other nuclei there can be no doubt of its occurrence, especially when sections such as that shown in Figure 8 (Plate **1**) exhibit precisely the same arrangement of the elements. In Figure 22 (Plate **2**) all the nuclei of each of the three divisions of the eye can be seen, though the eye as a whole has been somewhat distorted by pressure.

In each of the lateral eyes there are nine cells. This has been determined by careful reconstruction drawings from series of cross, sagittal, and frontal sections, and by counting the nuclei in entire preparations.

The number is invariable. Grenacher ('**79**, p. 64) found that there were eight cells in the lateral eyes of Eucalanus attenuatus, but there can be no doubt that in E. elongatus the number of cells is nine.

These cells are always arranged in a definite way, viz., in three superposed layers, a dorsal, a middle and a ventral layer, three cells in each layer (Plate **1**, Fig. 1). This arrangement is most easily determined by focusing upon the nuclei in entire preparations, as it is impossible to follow the cell walls throughout their extent. In this way one can readily see that there are three nuclei at a very high focus, if the eye is viewed entire from the dorsal side. The nuclei in this stratum are not at precisely the same level; the single one nearest the inner wall of the cup (Plate **1**, Fig. 10; Plate **2**, Fig. 22) is at the highest level; this and the other two which belong to the same group are represented in the same deep tone. It should be stated that in the figures mentioned the three nuclei in a given group have received the same tint, but this should not be taken to mean that they lie at precisely the same level in the eye. In all the groups of nuclei, the same arrangement may be observed; that is, there is one nucleus nearer the median face of the eye and two which are more lateral, though in the middle group the nuclei are much more nearly in the same antero-posterior line than in either of the other groups. The cell to which the median nucleus of the upper group belongs, is triangular in outline, and does not extend to the lateral border of the eye, while the remaining cells of the group are elongated and extend from the inner or basal portion of the eye to its outer margin (Plate **2**, Fig. 22). The same may be said of the remaining groups of nuclei also; the median cell is triangular and does not extend to the exterior, while the anterior and posterior cells reach from the base to the outer margin of the eye. It may be added that the three anterior and the three posterior cells converge somewhat from the inner (median) to the outer (lateral) face of the eye in the manner described by Grenacher ('**79**, p. 64).

It may be said, then, that the eye in Eucalanus elongatus contains in all twenty-eight retinal cells. In Cyclops, according to Richard ('**91**, p. 207), there are from 8 to 12 elements in each "Crystalline sphere," while Hartog ('**88**, p. 33) states that the median eye possesses "about eight peripheral and one central bacillus" and the lateral eyes "at least 8 to 10 peripheral bacilli and three central ones." Claus ('**91**, p. 246) was unable to determine with certainty the number of retinal cells in the eye of Diaptomus. At the beginning of his work, he found at least six nuclei, but later was led to believe that the number of cells was still greater. In Pontella, a type with compound eyes, the investigations of Parker ('**91**, p. 81) have shown that there are two cone cells in each retina, in addition to eight retinular cells and "other nuclei which probably represent undifferentiated cells."

It is plain that the number of retinal elements is accurately known only in Eucalanus elongatus and E. attenuatus, and that in these species the number is definite and constant both as regards the paired and unpaired portion of the eye.

The nuclei of the retinal cells are spheroidal or ovoidal and contain a chromatin network. The nuclei of other cells which appear around the eye (such as muscle cells, the hypodermis cells, and sheath cells) may be distinguished at a glance from those of the retinal cells by the difference in staining reaction. This of course appears to best advantage in haematoxylin stains, but is also evident in Mallory's connective-tissue stain. Figure 16 (Plate **2**) shows the differences accurately, and Figure 23 (Plate **2**) more diagrammatically. It will be seen that the nuclei of the retinal cells invariably contain more chromatin and so stain more deeply. This is an important character, for otherwise the nuclei of muscle or connective-tissue cells would be confused with them. It may be noted, also, that aside from their location, the nuclei of the optic cells are indistinguishable from those of the cells in the brain.

e. *Interior bodies* (*Phaosomes*); *their Arrangement in the Cells.*— The nuclei, however, are not the most noticeable bodies within the retinal cells. This characteristic belongs to the "interior bodies" (Binnenkörper), which Hesse (:**01**, p. 350) was the first to describe. He found that the interior bodies lie between the nucleus and the particular structure (Stiftchensaum) which, in his opinion represents the ending of the retinal nerves, but nearer the nucleus. These bodies have a greater affinity for stains, as Hesse has stated, than does the cell plasma. Hesse considers that they are essentially ribbon-like bodies, which may be so sharply bent or twisted that they seem to be divided into separate pieces. He thinks that branching of the bodies, if it occurs at all, is rare. He failed to find these bodies in Eucalanus attenuatus, but they are, he states, of constant occurrence in Calanus gracilis. He is unwilling to commit himself as to the part the interior bodies play in the reception of light, but states that they are not necessary for that function because they are absent from the retinal cells of Eucalanus attenuatus.

The observations of Hesse upon the median eye were made primarily to determine the method of nerve termination in the retinal cells, and he has given scarcely any attention to the interior bodies.

I can confirm his statements as to their position in general; but the bodies also occur almost at random in the cell, for in any series of sections it may be seen that they may lie between the nucleus and basal plates, or peripheral to the nucleus, as well as around it. It is true that they are found most numerously between the basal plate (probably the region of Hesse's "Stiftchensaum") and the nucleus. That this statement is correct for the lateral eyes, will be readily seen whatever the plane of section, and for the ventral eye in both sagittal (Plate **5**, Fig. 46; Plate **1**, Figs. 2, 3, *pha'so.*) and transverse (Plate **5**, Fig. 50) sections. Frontal sections of the ventral eye (Plate **1**, Fig. 8) show that the interior bodies are more numerous in the region between the nucleus and the median plane of the eye, than in the outer portions. But in any case the interior bodies are found less frequently lateral to the nucleus of a cell. This is shown in Figure 8 and appears also in all other regions of the eye.

I have been unable to confirm Hesse's statement that the rod-like bodies which appear to be isolated are sections of a ribbon-like or band-like, structure. My belief is that the interior bodies are really and only rod-like, or spicule-like, though ribbon-like bands are seen occasionally. One of these is shown in the right cell of the uppermost pair of Figure 8; but they are so infrequent and the appearance is so confused that the conclusion is almost forced upon one that in such cases they are so crowded together that their actual structure is not seen. It is easy to recognize the interior bodies in entire preparations of the eye, and I have never seen a band-like body in any preparations of that kind.

On the other hand, it is possible, if one follows many series, to find transitional forms between such closely aggregated groups of spicules as appear in Figure 8, and the ribbon-like forms. In Figure 2 (*pha'so.*) are shown several cases where it is plain that the structures which appear to be bands are in reality made up of numbers of single rods closely gathered together. The bands appear most frequently in material either stained deeply in vom Rath's fluid or very lightly in haematoxylin; in the one case the real structure is hidden by excess of stain, and in the other it is not brought out. I think that the non-appearance of bands in whole preparations almost precludes the possibility that these bodies are bands, and there can be no doubt at all that interior bodies occur as rods or spicules completely isolated from all other structures of the same sort. It must be admitted, however, that what seem to be band-like structures really appear in the retinal cells, as Hesse (**:01**) has stated and shown in his figure

(Fig. 1). The only question connected with their occurrence is whether or not that is their normal form. There can be no doubt that the rod-like forms are not optical sections or isolated portions of band-shaped bodies which have been so crumpled or folded as to be cut into many scattered bits. There is the possibility, though I believe it is remote, that both rod-like and band-like interior bodies occur in the retinal cells.

The question as to the branching of the interior bodies, which was raised by Hesse, is a debatable one. My belief is that branching does not occur, though in some preparations it appears to (Plate **1**, Fig. 5). Here, again, it seems probable that the branched appearance is due to the fact that numbers of rod-like bodies are closely massed together, the apparent branching being in reality due to the protrusion of the ends of certain spicules or groups of spicules from the more general aggregation of them. Indeed, the appearance of a branching habit seems to me to be strong evidence that the band-form of the interior body is really not the unit of structure in cases where it occurs, for often (Plate **1**, Fig. 8) an end of an otherwise apparently homogeneous ribbon is frayed out. I am at a loss to account for this frayed or tasseled structure except on the assumption that masses or groups of rods or spicules compose the ribbon.

The interior bodies stain in the same way that the nuclei do. This holds for all of the staining methods I have employed. The coloration is the same in the two objects in either iron-haematoxylin, Ehrlich's haematoxylin or acid haemalum. Vom Rath's fluid blackens or browns all the structures in the cell,— plasma, nucleus, and interior bodies, alike. But Mallory's connective-tissue stain is especially good for differential staining. When sections are treated in this way, the nuclei and interior bodies become yellow, all other structures red or reddish-purple, except chitin and connective tissue, which become blue. The interior bodies are evidently highly differentiated portions of the cell, though there is no reason to think that they are in any way the equivalents of nuclear structures, since their appearance in rather heavily stained haematoxylin preparations is vitreous and refractive. This is true to some extent, also, in Mallory's stain, and is especially evident if a vom Rath preparation is well decolorized in hydrogen peroxide. The interior bodies are ordinarily as deeply stained as any other part of the retinal cells, but they decolorize more quickly and then appear as refractive or colorless objects in the brown cytoplasm.

It is difficult to describe the interior bodies in general terms. The isolated ones are rod-like or spindle-shaped, and this is in general the character of the individual bodies in the masses which frequently occur. But other more or less irregular shapes may be seen, and are shown in the most of my figures. But here, as in the case of the bands or ribbons, I believe that the identity of the rod-like bodies is either lost by dense staining or not revealed because of too light staining.

The adequate illustration of the interior bodies in a cell is impossible. In all cases where they are shown they have been drawn as accurately as possible, but in no instance has any attempt been made to show all the bodies that are present. More attention has been paid to giving an accurate idea of the general appearance of a preparation, than to showing every detail in regard to the interior bodies.

The interior bodies in themselves appear to be homogeneous and structureless; no preparations that I have show them to be more than that.

But the shape and structure of the interior bodies — the questions as to whether the units are rod-like, or band-shaped, or both — are subordinate in importance to the arrangement of the bodies in the retinal cells, and the relation of this arrangement both to the direction of the nerve fibres in the retinal cells, and to the axes of the optic cups. In order to put this matter as clearly as possible, it will be necessary to describe the position of the optic cups, both with reference to the body of the animal and to each other.

The median, longitudinal axis of the ventral eye, as well as the optic nerve, lies exactly in the sagittal plane of the body, and the transverse axes are perpendicular to that plane.

The long axes of all the cells of the median ocellus of the eye are perpendicular to the median plane of the body (Plate **1**, Fig. 8). It is difficult to describe the axes of the central cell in this part of the eye, for the cell is quadrangular as seen in frontal section (Plate **1**, Fig. 8), the sides being of about equal length. But in Figure 8 the longer axis is parallel to the long axes of the remaining cells. The dorso-ventral dimension of the cells in the unpaired eye is approximately one-half that of their longest axis.

The chief axis of each lateral cup is a line perpendicular to the median side of the cup at its middle point. As is shown in Figures 7 and 9 (Plate **1**), such a line makes an angle of 45° with the sagittal plane, and with the dorso-ventral axis of the ventral eye, all three axes lying in the same transverse plane. In Figure 7 the cell which lies at the middle of the inner wall of the lateral eyes is the inner cell of the median group of three already described, and its long axis almost coincides with the chief axis of the eye. The cells which adjoin the central one, above and below, are, respectively the central cell of the upper and lower groups; the long axes of these cells are nearly at right angles to each other. The long axis of the dorsal cell is parallel to the dorso-ventral axis of the median eye, while the axis of the ventral cell is perpendicular to the median plane of the body. Similar relations are shown in the cross sections of the eye represented in Figure 9 (Plate **1**) and Figure 23 (Plate **2**).

An examination of frontal sections (Plate **1**, Figs. 2, 5) through the lateral eyes, or of entire preparations (Plate **1**, Fig. 10, Plate **2**, Fig. 22) gives an idea of the position of the axes of some of the other cells of the lateral eyes. It will be seen in Figures 2 and 5 (Plate **1**) that the axes of the peripheral cells of a group if prolonged would meet at an angle of about 90° at their outer ends. In Figure 2 the three nuclei shown in each of the lateral eyes belong to the cells of the median group of three. The central cell is seen to be triangular in outline, and the anterior and posterior cells are elongated. In Figure 22 (Plate **2**) the outlines of the cells in the right lateral eye are shown, but merely in the most general way, for it is impossible to indicate the cell walls in perspective, and even if it were attempted, the result would be confusing. But the figure shows well enough the relations that the axes of the anterior and posterior cells of the groups bear to each other, and it also indicates the general shape of the various cells, the central ones of each group being triangular, the others more elongated and extending from the base of the cup to its outer margin.

With the relations of the axes of the cells of the lateral eyes in mind, inspection of such a cross section as is shown in Figure 7 (Plate **1**) will convince one that the interior bodies have an arrangement that corresponds, at least in a general way, with the long axis of the cell. It will be seen that in the three cells forming in this section the median border of the lateral eyes these bodies are so arranged that they lie lengthwise of the cells. Not all the interior bodies have been represented in this drawing, but special care has been taken not to select those only which would prove the point here contended for. The illustration gives an accurate general idea of the arrangement of the rod-like or spindle-like bodies in the central cells. Even in such a seemingly heterogeneous assemblage of interior bodies as is shown in the lateral eyes in Figure 9 (Plate **1**), a definite arrangement with reference to the cells is apparent; and the same may be said of Figure 23 (Plate **2**).

In frontal sections a similar arrangement of the interior bodies with reference to the long axes of the cells is shown to exist. Figure 5 (Plate **1**) gives a fair general idea of a frontal section taken at a plane about midway between the nuclei of the median cells of the dorsal and middle groups. Even with the irregular and ragged appearance of the interior bodies (*pha'so.*) in the cells at the anterior edge of the eye, it is plain enough that they have a definite orientation in relation to the long cell-axes. Likewise, in the central cell there is a similar arrangement of the rod-like structures, which lie in a direction parallel to the line joining the anterior and posterior angles of the cell outline. Inspection of Figure 2 will show that in this case, also, the conditions are similar.

In the ventral eye, frontal sections give the best idea of the arrangement of the interior bodies in the retinal cells. Figure 8 (Plate **1**) shows, in its general features, the condition that appears in all cases. The interior bodies are arranged with reference to the long axes of the cells, except in the case of the anterior paired cells. This arrangement is least apparent in the central retinal cell, but this is the cell in which the "sides" are more nearly equal than in any other. Sagittal (Plate **1**, Fig. 6) and cross sections (Plate **1**, Fig. 7; Plate **2**, Fig. 23) show that there is a somewhat similar disposition of the interior bodies as regards the dorso-ventral axes of the cells.

In preparations of the entire eye, the interior bodies may be seen to have the same arrangement that is shown in sections. Figure 1 shows this in a rather vague way for the lateral eyes. But in Figure 21 (Plate **2**), where the representation of the interior bodies is believed to be accurate, as far as they are shown, their arrangement in the ventral eye is much like that shown in Figure 8 (Plate **1**), particularly as regards the anterior cell. In the other cells of the eye, especially the paired ones, the arrangement corresponds in a general way to that in Figure 8. All the interior bodies that could be seen in the retinal cells of the *ventral eye* have been shown in black.

The definite arrangement of the interior bodies with reference to the long axis of the cells appears in general, and in every cell, in any preparation. But if the attempt is made to discover whether every interior body is so arranged, it will be seen that there are many exceptions; and some such appear in all of the figures. It is possible that such exceptions may be more apparent than real, owing to the plane in which a section is cut, but they are none the less difficult of explanation. There can be no doubt, however, as to the general facts.

The correspondence in arrangement that appears in cross and sagittal sections of the eyes, leads one to question whether the interior bodies, considered in their spatial relations, are not really plate- or disc-like. It seems to me that this is possibly the case, though I have been unable to satisfy myself completely of the correctness of this view by following them on serial sections. And I can not see that in entire preparations of the eye the interior bodies have the form of plates. On the contrary, they always appear to be rod- or spindle-shaped. If the bodies are not discs or plates, it seems that the other alternative which will account for their positions in the cell is to consider that some of the rod-like forms are parallel to one axis of the cell, and some to another axis. For example, in comparing the central cells in Figures 5 and 7 (Plate **1**), the one seen in frontal, the other in cross section, it is plain that in each case there are interior bodies which are arranged with reference to the longer of the two axes of the cell which appear in each of the two figures.

Structures which seem to be of a nature similar to the interior bodies of Eucalanus, have been described for other Copepoda and lower Crustacea. Hartog ('**88**, p. 34) states that there is an "oblong body (probably a rhabdome) staining deeply with osmic acid," "in the inner limb of each bacillus," and Claus ('**91**) has mentioned the general occurrence of "Cuticular-stäbchen" in many other crustacea possessing a median eye, as well as in Diaptomus, Anomalocera, Pontellina and others among the Copepoda. One can not fail to note a certain similarity between the condition he figures in the dorsal eyes of Pontellina mediterranea (Claus, '**91**, Taf. iv, Fig. 6–8) and those in Eucalanus. He states (p. 350) that the cuticular rods were intensely colored in borax carmine or haematoxylin, and were not straight, but curved, and joined in pairs at their thickened ends. Parker ('**91**, p. 81) says that in the portion of the retina surrounding the cone in Pontella "the most conspicuous structures . . . . are rod-like bodies, which probably represent rhabdomeres." He detected eight such bodies, and believes that there is a cell for each rod, because a large nucleus is near each of the latter. Consequently it would seem, that, if the rhabdomeres and "Cuticular-stäbchen" are homologous with the interior bodies, the numbers do not nearly correspond in the various cases where the structures occur. There is certainly more than one interior body for each nucleus in the retina of Eucalanus. Hesse (:**01**, p. 351) mentions the similarity of the structure in the retinal cells of Eucalanus with the interior bodies in the visual cells of the leeches and in the problematical visual cells of the Lumbricidae.

f. *Relation of Axis Cylinders to Retinal Cells.*— That the interior bodies in the cells of the eye of Eucalanus have a functional importance, seems to me to be indicated by the innervation of the retina. The facts concerning the latter have been rather confused, if one considers all the accounts in the literature. In general the manner of innervation has been looked upon as a factor in determining the phylogenetic relationship of the Copepoda.

Grenacher was the first to determine the way in which the axis cylinders are related to the retinal cells. He says (Grenacher, **'79**, p. 65) that the fibres of the optic nerve "are continuous with the inner, pointed ends of the cells," meaning the ends nearer the pigment plate. He was able to follow the nerve fibres into the cells.

Following Grenacher's work, came that of Hartog (**'88**, p. 33), who found that in Cyclops the ocellus receives the nerve posteriorly at the outer surface, so that the optic elements are reversed as in the flatworm Dendrocoelum. The partition which separates the eye from the brain is "quite imperforate by nerves." Further on (p. 34) he states that in both Cyclops and Calanus he followed a few fibres along the septum between the blocks of the lateral ocelli, and had positive evidence that such do not enter the "bacilli"; they may end in the nuclei of the blocks or pass on to the frontal region. And in a footnote (p. 34) he describes the results of dissecting and sectioning the eye of Calanus in alcoholic specimens; he there found that "the lateral branches [of the optic nerve] unquestionably do not enter the inner ends of the bacilli"; he was unable to speak with certainty about the ventral eye.

As already stated, Claus (**'91**) did not confirm Grenacher's observations as to the point at which the optic nerves leave the cells of the eye. Claus was evidently of opinion that the nerve leaves from the outer side of the visual cells and that the recipient ends of the fibrils are turned toward the pigment body, thus making the median eye an "inverses Becherauge." He found this to be the case in the Cypridinidae, Branchiopoda, Cladocera and Argulidae and implies that it is so in the Copepoda and Cirripedia.

Richard (**'91**, p. 208) also found that the retinal elements are "renversés comme dans les yeux marginaux des Pecten et dans celui des Vertebrés," for the optic nerves do not enter the pigment mass, but pass to the dorsal margin of each simple eye and terminate at the surface.

Other investigators are more or less inclined to consider the parts of the median eye as inverted. Schmeil (**'97**, p. 30) accepts the statement that such is the case. Carrière (**'85**, p. 178) leaves the question open, but considers the resemblance of the eyes of Calanella (Eucalanus) to those of Clepsine or Planaria "as unmistakable." Lang

('**88**–**94**, p. 361), also, uses practically the same expression, though he considers it as unsettled whether the nerves leave the cells from the inner or the outer ends. And Hesse (:**01** and :**02**) states many times that the median eyes of Crustacea are inverted.

As far as my observations on Eucalanus elongatus go, I have been able to confirm in every respect the statements of Grenacher ('**79**) concerning the relation of the optic nerves to the retinal cells.

As already stated, the bundle of fibres (axis cylinders) composing the optic nerve leaves the eye directly at its posterior border, dorsal to the basal plate of the ventral eye and between the posterior basal plates of the lateral eyes. This may be readily determined from whole preparations (Fig. 1, *n. opt.*), and is seen in sagittal sections (Plate **1**, Fig. 6, *n. opt*; Plate **5**, Figs. 44, 46, 48) and frontal sections, though more clearly in the former. If the section coincides precisely with the sagittal plane, the basal plates of the lateral eyes are not cut, but the relation described above will appear in cross (Plate **5**, Fig. 49) or frontal sections (Plate **1**, Figs. 2, 5). Immediately behind the eye (Plate **4**, Figs. 38, 39) the optic nerve is circular in cross section, but within the eye it is separated into parts, which have come from the two lateral portions and the single and ventral portion of the eye (Plate **1**, Figs. 2, 5, 6).

g. *Numerical Relation of Nerve Fibres and Visual Cells.*— The number of fibres in the optic nerve corresponds precisely with the number of retinal cells in the eye as a whole. This can be so readily seen in cross sections of the nerve (Plate **4**, Figs. 38–43) and has been observed in so many cases that there can be no doubt on that point. In the region of the optic nerve behind the eye before the frontal nerves have joined it (Plate **1**, Fig. 1; Plate **4**, Fig. 38) the fibres are closely massed into a single cylindrical bundle, but posterior to this region the fibres gradually become more widely separated from one another (Plate **4**, Fig. 39; Plate **5**, Fig. 47). About half way between the eye and the brain and thence to the brain, the cross section of the nerve is very much flattened dorso-ventrally and elongated laterally (Plate **4**, Figs. 40, 41). In any cross section of the nerve posterior to the point at which the frontal nerves (*n. f.*) join it, the latter are always distinguishable by the presence of a fibre which has a delicate sheath staining black in vom Rath's fluid (Plate **4**).

Near the eye, the fibres of the optic nerve are rounded in cross section, and each is provided with a delicate sheath, which, though distinguishable from the axial bundle, is very closely applied to it (Plate **4**, Figs. 38, 39). But farther from the eye, the sheath and the central strand become separated (Plate **1**, Figs. 40–43). If is difficult to say whether this is due to the action of the reagents or whether it is natural. In Figure 40 (Plate **4**) some of the fibres are seen to lie in clear spaces within the sheaths while in some the sheath and central portion are everywhere in contact. The outline of the sheaths alone in the former case is about as extensive as the entire structure in the latter case, and the central part is much smaller in the fibres where it is not in contact with the sheath. This would lead one to think that shrinkage of the central bundle had occurred; but in more posterior sections of the same series (Plate **4**, Fig. 41) there are more fibres which are separated from the sheaths by a space.

On the other hand, in Figures 42 and 43 (Plate **4**), which are from another series, and pass through the anterior part of the brain, the axial strands are all about equal in size, but each is separated from its sheath, the extent of the separation varying in different cases. And in Figure 54 (Plate **5**), which is from a section whose plane is farther back in the brain than that represented in Figure 43 (Plate **4**), some of the fibres are distinctly separated from the sheaths, while in the others the sheath could not be seen. But in this case all the central strands are of approximately the same size. If shrinkage occurred at all, one would certainly expect it to affect the nerves at points near the eye as well as farther back, but I have never observed this in any series.

It is evident from what has just preceded that the individual fibres of the optic nerve preserve their identity from the retinal cells to the brain. Figures 42, 43 (Plate **4**), and Figure 54 (Plate **5**) show that twenty-eight fibres may be distinguished some distance posterior to the point where the nerve enters the brain as readily as immediately behind the eye. Figure 54 (Plate **5**) is from a section at about the level where the individual fibres in that series are no longer distinguishable.

The fact that there are twenty-eight cells in the eye and the same number of fibres in the optic nerve is strong *a priori* evidence that there is one cell for each fibre, and if the distribution of the fibres to the retinal cells is followed, one can hardly fail to be convinced that such is the case. Sections stained by Mallory's connective-tissue method (Plate **5**) are especially favorable for tracing the fibres. It is hardly necessary to state that not all preparations are equally valuable and that even when the staining has been particularly successful in differentiating the nerves from other tissue, it is not easy to make out the course of the nerves. Furthermore, if in any case the plane of the section is unfavorable, it is practically impossible to trace the individual nerves.

As previously stated, the optic nerve leaves the central (or pigment) cell of the eye at its posterior border, the axis cylinders having passed through that cell in their course from the visual cells of the eye. The fibres can enter the pigment cell only at the points where the optic cups are in contact with the central cell. Figure 2 (Plate **1**) shows that the lateral optic vesicles are separated from the tapetal layer of the pigment cell everywhere (at the level of the section) except at the anterior angle of the anterior basal plate and at the posterior angle of the posterior plate, and that in these places the tapetum is interrupted. In cross sections (Plate **5**, Figs. 49, 50), it can be seen that the lateral eyes (Plate **5**, Fig. 49) are in contact with the central cell (*cl. c*) at the dorsal and outer ventral margins of the latter; and in serial cross sections it is shown that the eyes touch the central cell along its entire dorsal and ventral margins, for in no section of such series are these connections broken. But it should be said that whereever the lateral eyes and central cell come in contact, the surrounding capsular membrane of the eye vesicle is present. I believe that the spaces separating the eyes from the central cell (Plate **1**, Fig. 2; Plate **4**, Figs. 49, 50) are not artifacts due to shrinking, because they occur in every preparation, no matter what the treatment may have been, and in all preparations the conditions are precisely as described. Again, it seems reasonable to maintain that, if the spaces were due to shrinkage, there would be evidence to this effect in wrinkling or irregularities of outline, but this is not the case.

In the ventral eye, the relations of the central cell and optic cup are similar to those described for the lateral eyes. Figures 6 (Plate **1**), 44 and 48 (Plate **5**) show that the two are in contact at the anterior and posterior margins of the eye and also at a point midway between the two. As in the paired eyes, the tapetum (*tap.*) is interrupted where the eye touches the pigment cell (*cl. c*). These anterior and posterior regions of contact also mark the anterior and posterior limits of the basal plate of the eye, but the other place of contact is through an opening in the basal plate, the plate being continuous lateral to the region of contact, as it is anterior and posterior to it. (Compare Plate **1**, Fig. 6, and Plate **5**, Figs. 49 and 50.) The ventral component and the pigment cell (Plate **5**, Fig. 49) are also in contact lateral to the basal plate (*la. ba.*) of the ocellus and nerves leave the retinal cells at these points.

As a basis for describing the nerve supply of the eye, we may take

Figure 6 (Plate **1**) and compare with it other sagittal sections (Plate **5**, Figs. 44, 46, 48) as well as cross sections (Plate **5**, Figs. 45, 49, 50). In Figure 6 (Plate **1**) are shown the ventral eye, at the right, in contact with the central cell (*cl. c.*) at three places, and the optic nerve (*n. opt.*) leaving the eye as a single bundle of fibres, which results from the union of three divisions lying in the median plane of the whole eye, between the lateral cups. The dorsal division passes through the central cell, between the paired ocelli, from the anterior border of the cell, and is shown in greater extent in Plate **5**, Figure 44. The ventral and posterior of the three median divisions of the optic nerve (in Plate **1**, Fig. 6) leaves the ventral eye at the posterior end, between the basal plate and the bounding membrane of the eye (compare Plate **5**, Fig. 46). As regards the middle bundle, the way in which it leaves the ventral eye is shown in the sagittal sections in Figures 6 (Plate **1**), and 44, 46, 48 (Plate **5**), with which the cross section shown in Figure 50 (Plate **5**) should be compared. Figures 6 and 50 show particularly well that the nerves which leave the central portion of the ventral eye pass through an opening in the basal plate. Frontal sections of the eyes show that there are lateral, as well as median, divisions of the optic nerve (Plate **1**, Fig. 2).

In the cross-sections represented in Figures 45, 47, and 49, which are three of a series of 8 sections, and are successive (Fig. 47 being posterior and Fig. 49 anterior), the same conditions are encountered. In Figure 45 (Plate **5**) are shown 28 axis cylinders (*fbr.*) in approximate cross section. There are five in each lateral division, two leaving the ventral eye, and sixteen others are in a median group though their departure from the retinal cells is not shown in the section. In the section anterior to this (Plate **5**, Fig. 49) there are thirteen fibres in the central cell: of the five lateral ones shown in Figure 45 the distribution of those which are more dorsal cannot be followed, though that of the remaining four is shown in relation to the left lateral ocellus (*oc. s.*). Three fibres leave the lateral eye and one (on the same side) leaves the ventral eye. The cell of the ventral eye shown on the right is the left cell of the second pair from the posterior end of the eye (cf. Plate **1**, Fig. 8). It will be noted that the fibres in this section, which are distributed to the ventral eye, pass lateral to (right and left of) the basal plate instead of through it.

In the next section anterior, (Fig. 50) six fibres are shown which are still in the pigment cell, while two others diverge toward the lateral eyes, and one more is barely distinguishable in the ventral portion of each of the lateral eyes. In the ventral eye may be seen the central opening in the basal plate (*la. ba.*) and one nerve fibre (*fbr.*) passing through it. This fibre belongs to the anterior cell of the ventral eye and is shown again in Figures 44 and 48. It may be said here that this particular fibre is of constant occurrence and distribution, and more easily followed than any other one. In Figure 44 some other fibres of the ventral eye are shown, passing through the basal plate, though it is not possible to trace them from individual cells.

The fibres of the dorsal group shown in Figure 6 (Plate 1) pass through the pigment cell to the anterior border (Plate 5, Figs. 44, 48), where they diverge to the right and left. The cells to which these fibres belong are the anterior cells of the lateral ocelli.

It is difficult to follow the nerve fibres in frontal sections. There are a few cases, however, which agree with the conditions as described from sagittal and cross sections. In Figure 53 (Plate 5) are shown several fibres (*fbr.*) passing from the lateral eye of the right side. It is plain enough that they leave by the basal or inner ends of the cells, as already shown in many other cases.

Whether one believes that the central cell (as I have called it) or the basal plates contain the pigment of the eye, is immaterial in considering the nature of the innervation of the retinal cells, viz., whether the eye is of the inverted type or not. In any case it cannot be claimed that the nerves spread over the outer faces of the cells and there gain entrance to the cells themselves for distribution toward their basal ends. The contention of previous investigators with regard to the inverted character of the median eye has been based upon their observations that the nerve fibres *leave from the outer sides or ends of the cells.* But in the eye of Eucalanus there is not an instance where an axis-cylinder fibre leaves from any part of a visual cell except that which must be regarded as inner, or proximal. It might seem that the axis-cylinder which belongs to the anterior cell of the ventral eye (Plate 5, Figs. 44, 48) is an exception to the above statement, but even in this case the fibre passes through the basal plate and instead of leaving that particular cell at the distal end, really does so from the side which is directed toward the centre of the eye. The case of this cell certainly differs in some respects from any other that shows the relation between cell and fibre, but I believe it is a difference in degree and not in kind.

The only way by which the retinal cells in Eucalanus could possibly be regarded as "inverted," is to assume that the axis cylinders, which leave the cells at their basal ends, pass toward the distal ends without dividing into fibrillae, and that at the distal ends of the cells they turn back. In that event the nerve-endings would be directed toward the pigmented part of the eye, and the eye would necessarily be regarded as inverted. It need only be said that there is no evidence in my preparations that nerve fibres and retinal cells are thus related, though that is clearly the condition which Claus (**'91**), Hartog (**'88**) and Richard (**'91**) had in mind. Therefore, if the relation between the cells of the eye and the axis cylinders that arise from them is to be regarded as evidence that the eye is inverted or not,— that is, that the recipient portions of the nerve fibre are or are not directed toward the pigment and away from the exterior,— we must, at least in this case, look upon such evidence as distinctly against the view that the median eye in Copepoda is inverted.

h. *The Neurofibrillae.*— But more direct evidence than that hitherto presented is at hand relative to the character of the eye in Eucalanus; it is based upon the relations existing between the end-fibrillae of the nerves and the retinal cells. The character of the nerve ending in the visual cells of Eucalanus has been investigated by Hesse (**:01**, p. 349) in the course of his studies on the eyes of invertebrates. He states that he was led to this investigation by a desire to know whether or not the relationship believed to exist between the median eyes of Crustacea and the eyes of flatworms, did not also extend to the "lichtrecipierenden Theile der Sehzellen." He had previously shown (Hesse, **'97**) that in Planaria and its allies there is in the visual cells a "Stiftchensaum .... dessen einzelne Stiftchen nichts Anderes sind als verdickte und vielleicht stofflich etwas veränderte Enden von Neurofibrillen, welche von der Nervenfaser in die Sehzellen einstrahlen" (Hesse **:01**, p. 350). With regard to the Copepoda he says (**:01**, p. 350): "Die Untersuchung hatte das vermuthete Ergebnis: ich fand, dass das 'Stäbchen' bei diesen Formen ein Stiftchensaum ist." His preparations of Eucalanus elongatus showed this condition the most clearly, although the conditions in Eucalanus attenuatus and in Calanus gracilis were practically the same.

I have been unable to confirm, by my preparations, Hesse's statements with regard to the presence of a Stiftchensaum in Eucalanus elongatus. The iron-haematoxylin method following corrosive-acetic fixation (which Hesse seems to have employed) is the least successful of any that I have used in showing even the general relations between axis cylinders and retinal cells.

But any preparation will show in the basal region of the retinal cells an almost indistinguishable striation of the cytoplasm. This striated appearance is seen to best advantage in vom Rath preparations (Plate **1**, Figs. 5, 7, 9), though it is shown to some extent in sections stained with iron-haematoxylin (Plate **2**, Fig. 23), or in Mallory's connective-tissue stain (Plate **5**, Figs. 44, 50). Even the granules of the cytoplasm take part in the general radiate arrangement. It is also very plain that the contents of the cells are more densely aggregated nearer the basal plates than toward their distal ends (Plate **5**, Fig. 50).

It is difficult to say whether this striation is in any way related to the "Streifung" which Hesse notes in connection with the "Stiftchensaum." But in my opinion the denser condition of the cell contents, as well as the striated appearance in the retinal cells, is an expression of the greater secretory activity of the cells in that region, the product of which is shown in the basal plates, since the latter must be regarded as products of the retinal cells, for there are no others from which they can reasonably be derived.

Such striations are strikingly shown, also, in the cells of the digestive tract in Eucalanus (Plate **4**, Fig. 52), where it is very unlikely that they can be regarded as due to the presence of end fibrillae of nerves spreading out into the cell contents. This condition of the digestive-cells, I believe, is strictly in line with that shown by Mark('**76**) to exist in the cells of the salivary glands of certain Coccidae.

The true character of the neurofibrillae is shown in such a drawing as Figure 49 (Plate **5**), where, in the cells of the ventral eye, the structures that I take to be the neurofibrillae of the axis cylinders are shown as rather heavy, beaded bodies (*n. fbrl.*). These lie at a very low focus in the section (a fact which cannot be expressed in the drawing), and are in all likelihood parts of the two nerve fibres shown in a corresponding position in Figure 45. In the left lateral eyes (Fig. 49, *ocl. s.*) there are two nerve fibres which may be seen to be directly connected with similar structures. In the right eye one such is shown. The nerve endings when shown in their entire extent (as I believe is the case in Figure 49, *n. fbrl.*) stand out with remarkable clearness from the rest of the cell, and they take a tint almost precisely the same as that of the nerve fibres outside the cells. It is not unlikely that some of the granular appearance in the basal parts of the cells of the eyes is due to the presence of the beaded nerve-terminations. This is indicated rather strongly in the right lateral ocellus shown in Figure 49.

As far as my observations go, therefore, the median eye of the Copepoda does not resemble the eye of the flatworms in having the recipient ends of the nerves turned toward the pigment, nor in the character of the nerve ending, since it does not possess a "Stiftchensaum" in the sense in which Hesse uses the word. It might be said here that Nowikoff (:**05**, p. 449, 450) has failed to confirm the observations of Hesse (:**01**) as to the presence of a Stiftchensaum in the median eye of Branchipus. It seems to me probable that the Stiftchensaum which Hesse (:**01**, Taf. 16, Fig. 1, *a, sti.*) has shown is in some way a part of one of the basal plates. The general outline of the area covered by the "Stiftchen" in his figure resembles closely sections of the basal plates of the lateral eyes in the form I have studied (Plate **1**, Fig. 2, *la. ba.*). However, all the evidence of striation that I have seen is invariably distal to the margin of the plates. And the latter seem to me to be as nearly homogeneous as it is possible for an object to be.

i. *Relation between Neurofibrils and Interior Bodies.*— It has not been possible to discover, with certainty, a *direct* structural relation between the interior bodies and the nerves, though there is a strong indication that a functional relation exists between them. There is, indeed, some evidence that the nerve fibres and interior bodies are *continuous.* In the whole preparation shown in Plate **2**, Figure 21, there can be no doubt that the blackened structure in the opening of the basal plate of the ventral eye, in the centre of the drawing, is the group of nerve fibres which may be seen in many sections to leave the eye at that point (cf. Plate **1**, Fig. 6). In the case under consideration (Fig. 21) it is very plain that there is a direct connection between the nerves and one of the interior bodies, and I cannot draw the line between what should be called interior body, and that which is strictly nerve fibre. There can be no doubt of the fact that the two are not distinct structures in the sense of their being separated from each other.

In sections, also, the indications are strongly toward a structural, and therefore presumably functional, relation between nerve fibrils and interior bodies. Each of the former ends distally in a club-shaped enlargement (Plate **5**, Fig. 49), and this is often, if not always, in very close apposition to a group of interior bodies. The latter generally lie in a clear space in the cytoplasm of the cell, and it is difficult to find any other structures within the space. But in Figure 49 in the right cell of the ventral eye (left in the figure) is shown a nerve fibril (the one nearest the median plane) which, at its peripheral end is divided into two twigs, each bearing a club-shaped enlargement; and between the enlargements there lies an interior body. It is certain in this case that the enlargements of the nerve-fibrils are within the vacuole which contains the interior body. Various other, though similar, conditions are shown in Figures 44, 46, and 48. In many cases where a nerve fibril cannot be followed continuously throughout the cell, the enlargements at the ends may be seen around the margins of the spaces (vacuoles) surrounding the interior bodies (Plate **5**, Fig. 50). For a preparation stained in vom Rath's mixture, Figure 17 (Plate **2**) is extremely suggestive of a continuity between nerve fibres and interior bodies, though such a continuity can not be directly seen.

The arrangement of the interior bodies with regard to the long axes of the cells corresponds, in general, to the course of the nerve fibrils in the cells. This is shown in Figures 49 and 50 (Plate **5**). Furthermore, the nerves which in Figures 2 and 5 (Plate **1**) are shown continuing between the lateral eyes would, as fibrils within the cells, have the general direction of the isolated interior bodies, or the long rows of them (cf. Plate **5**, Figs. 46 and 48 with Plate **1**, Figs. 2 and 5).

It seems to me that these facts all point strongly toward an intimate structural relationship between the interior bodies and the nerves of the eye, and certainly suggest that the former are functional parts of the visual cells as such.

j. *Parts of the Eye in their Relation to the Hypodermis.*— So far we have considered the median eye merely in its anatomical relations without making a comparison between it as a type and other characteristic eyes of Crustacea. Parker ('**91**, p. 47) states that in Crustacea "at least three types of retinal structure can be distinguished," depending upon the final form assumed by the retina in its development from a simple "thickening in the superficial ectoderm." The first type is found in Decapoda, Schizopoda, Stomatopoda, Isopoda, Nebalia and the Branchiopodidae, and is merely a thickening of the hypodermis, which retains a superficial position permanently. The retina is directly continuous at its edges with the hypodermis.

The second type is found in the Apusidae, Estheridae and Cladocera. This type is distinguished from the first by the fact that the retina comes to lie beneath a fold of integument, instead of remaining permanently at the surface of the body. In the Estheridae, as represented by Limnadia, the eye lies in a pocket which communicates with the exterior by means of a pore. In the Cladocera this pocket becomes closed and partly obliterated, so that the retina is then not continuous with the ectoderm. The right and left retinas may remain separate (Apusidae), lie close together (Estheridae), or fuse (Cladocera). The three groups in which the second type of retinal structure is found represented form a natural series, beginning with the Apusidae and extending through the Cladocera.

The third type of retinal structure is found in Amphipods and "possibly in Copepods." The essential point is that the retina is separated from the hypodermis, but not by the formation of an optic pocket. The method of separation in Amphipods is by means of a membrane, the corneo-conal membrane, which is, in Parker's opinion, composed of two layers, one formed by the retina, the other by the hypodermis. The two portions of the corneo-conal membrane are seen to be separate at the edge of the retina, one being the basement membrane of the hypodermis, the other forming the capsular membrane, which envelopes the retina and is finally reflected over the optic nerve. In Gammarus the corneo-conal and capsular membrane completely enclose the retina and separate it from all other tissues with the exception of the optic nerve.

In Copepoda, too, the retina is separate from the hypodermis, and in Argulus the separation is made more extensive by an intervening blood space. The retina in the Eucopepoda as represented by the Pontellidae and Corycaeidae is apparently not continuous with the hypodermis, but Parker (**'91**, p. 59) states that it is difficult to decide to which of the three types the retina in Copepoda belongs. ". . . . If the lateral eyes in Copepods are not representatives of a fourth type, essentially different from the three already described, they must be considered members of the third retinal type." Hesse (**:02**) has grouped the eyes of many invertebrates in a way similar to that employed by Parker (**'91**), but the work of the former writer will be referred to more extensively later.

It may perhaps be questioned whether the median or "nauplius" eye of such Copepoda as Eucalanus, Cyclops or Diaptomus, may be justly compared with the eyes of a distinctly higher type, which are found in other Crustacea. But the relation of the parts of the eye to the ectoderm in Eucalanus are striking, and exactly along the lines marked out by Parker (**'91**) in his treatment of the compound eyes in Crustaceans. It seems to me, therefore, that it is worth while to consider the matter, since it is important in a discussion of the phylogeny of the median eye.

Nothing is known concerning the ultimate relation of the median eye to the hypodermis, or other membranes, except that it is developed from ectoderm (Grobben, **'81**; Urbanowicz, **'81**; Claus, **'91**, p. 259), as in all other Crustacea. Claus (**'91**, p. 260) treats of the enveloping membranes in the following words: "Endlich hat der mehr oder minder herabgerückte Augenbecher, und im Falle der Vereinigung der dreitheilige Augencomplex, eine mesodermale Umhüllung erhalten, welche sich direct in das Neurilemm des zur Retina tretenden Nerven fortsetzt." Grenacher ('79) makes a similar statement. Both Claus ('91) and Hesse (:01, :02), however, state that the median eye of Crustacea lies outside the ectoderm.

My preparations show that this statement quoted from Claus ('91) is only partly correct. The optic nerve is surrounded by a very distinct membrane, which becomes blue in Mallory's stain, and is therefore to be regarded as composed of connective tissue, and is probably of mesodermal origin. The membrane enveloping the optic nerve is continuous with the one around the brain (Plate 1, Fig. 4), but has nothing at all to do with the neurilemma of the nerve fibres (Plate 5, Fig. 47). But the discussion of the relation between this sheath of the optic nerve and the eye may be deferred for the present.

In its relation to the hypodermis, or ectoderm, the ventral part of the median eye corresponds in every way to the first of the retinal types mentioned by Parker ('91), though Hesse (:02) states that the whole median eye is detached from the ectoderm. Both cross and sagittal sections of the eye (Plate 1, Figs. 3, 6; Plate 5, Figs. 44, 48) show with perfect clearness that the ventral portion of the tripartite eye is merely a thickened region in the ectoderm (*h'drm.*), and has maintained its superficial position permanently. The hypodermis and retinal cells are adjacent; in other words, the retina and ectoderm are continuous. The basement membrane of the ectoderm may be traced in sagittal (Plate 5, Figs. 44, 48) and in cross sections (Figs. 49, 50) continuously from a region entirely outside the retina, over (dorsal to) the basal plate of the eye, and on to the ectoderm as such again. This basement membrane is very delicate, but visible with perfect clearness in any preparation, though Mallory's stain shows it most distinctly. In this stain the membrane does not become blue, and so should be regarded as of a different nature from that enveloping the optic nerve and brain.

Such nerves as leave the ventral eye through the basal plate, must penetrate the basement membrane of the hypodermis as well as the basal plate, since the plate is a product of the cells composing the retina, and these are plainly specialized ectodermal cells. The nerve fibres which in passing from the visual cells do not penetrate the basal plate pass through the basal membrane only.

The relations of the dorsal components of the eye and their enveloping membranes are not so easily made out as in the case of the ventral eye. Each of the paired eyes is surrounded by a delicate sheath (Plate 5, Figs. 46, 50), but in the adult condition it is evidently not related to the ectoderm as in the ventral eye. Such illustrations as

Figures 3 and 6 (Plate **1**) and 44 (Plate **5**) will make clear a condition that is constantly met with. It will be seen that at the anterior margin of the central cell there is plainly a separation of what appears (Plate **5**, Fig. 44) to be a single membrane at a little distance from the eye. One of the two portions of it evidently is dorsal and one ventral as regards the paired portion of the eye. It seems to me that no other interpretation of this condition is possible than to regard these membranes as belonging to the ectoderm, especially since in a number of cases the membrane, which is to all appearances single, becomes so closely applied to the ectoderm that the two are indistinguishable.

The sheath of the optic nerve may be traced in sagittal sections (Plate **5**, Figs. 44, 48, *mb. pi'n.*) for some distance over the dorsal surface of the eye, but on the ventral side of the nerve (in a sagittal section) it seems to be interrupted at the posterior margin of the central cell (Fig. 44). It appears to fuse with the basement membrane of the hypodermis at this point. I cannot say whether the membrane of the optic nerve is reflected over the hypodermis in a posterior direction or not, for it is impossible to distinguish the two. Likewise, I have been unable to find any evidence that the nerve-sheath passes forward over the basal plate of the ventral eye with the basement membrane of the hypodermis. In Figure 44 the membrane of the optic nerve is shown passing over the dorsal surface of the lateral eye for a short distance, but it cannot be followed over the whole eye. It is possibly continuous with the membrane that passes over the dorsal surface of the eye, and in front of the eye it seems to unite with another membrane from the ventral side of the paired portion of the eye (Plate **5**, Fig. 44; Plate **1**, Fig. 3). The resulting, apparently single, membrane shortly becomes indistinguishable from the hypodermal basement membrane; but if so it cannot be objectively identified by any differential staining reaction. The membrane of the optic nerve (*mb. pi'n.*) may also be seen in cross sections (Plate **5**, Figs. 45 and 47); in Figure 46 (Plate **5**) it (*mb. pi'n.*) is shown enveloping the lateral eye upon the sides, at least partly.

My belief is that the membrane around the optic nerve does not envelope the entire eye, as has been maintained by Grenacher ('**79**) and Claus ('**91**). All the direct evidence that I can acquire goes to show that it rests something like a cap over the posterior portion of the eye, not passing over the dorsal surface of the ventral eye at all. However, the relation of this membrane to the eye is not so important in a general consideration, as the relation that the parts of the eye bear to the body ectoderm; and the further discussion of this subject may therefore be deferred until later.

Thus far in this paper, I have described the median or tripartite eye of Eucalanus elongatus as a type of this structure among Copepoda. I have tried to show that none of the parts of the eye may be considered as inverted in the sense that the nerves leave the retinal cells from their distal ends. The nerves pass through the basal plates of the optic vesicles and, in part, traverse the entire extent of the "central cell" of the eye, on their way to the brain. It is immaterial, in considering the relation of optic fibres to sensory cells, whether the "central cell" or the basal plates are held to be the pigment bearers of the eye. The relations of axis cylinders and cells remains the same. There can be no reasonable doubt that there is one nerve fibre, and only one, to each retinal cell, for in several instances such a nerve fibre has been traced from its emergence from a single cell which gives rise to no other fibres; and, furthermore, the number of fibres in the optic nerve and the number of sensory cells in the eye is precisely the same in all cases. It also seems very probable that only the unpaired portion of the eye retains, in the adult, its original relation to the hypodermis; the lateral eyes are without doubt no longer directly continuous with the hypodermis as is the ventral eye. The foregoing are the principal facts which it will be necessary to consider in a general discussion.

2. The "Inverted" Eyes or "Organs of Claus."

It remains, now, to describe certain other structures which are probably optical in function. I shall call these "the organs of Claus," from their discoverer. Claus ('**63**, p. 56) was the first to describe the organs in question, and since that time no one has investigated them in any way, so far as I know. Richard ('**91**, p. 209) states that they are not found in Cyclops, and Hartog ('**88**, p. 33) located similar "concretions" at the base of the fifth feet in Cyclops brevicornis. Claus's observations were made upon Eucalanus attenuatus Dana (Calanella mediterranea); his description of the organs is as follows "Gehörorgane wurden nicht mit Sicherheit beobachtet, möglicher Weise aber gehört in die Kategorie dieser Organe eine eigenthümliche Bildung im Gehirnganglion von *Calanella.* Es sind zwei kugelige, Gehörblasen ähnliche Räume, in deren hellem Inhalte ein Ballen von Concretionen bemerkt wurde. Ob diese Differenzirung regelmässig auftritt oder nicht, habe ich leider unterlassen zu entscheiden."

a. *Location.*— The organs are symmetrically located within the brain at its anterior end (*o. Claus,* Plate **2**, Fig. 24; Plate **3**, Figs. 25, 27, 35; Plate **6**, Fig. 56). They are in the closest apposition with the great nerves (*n. at.*) which supply the first antennae, and they lie on the median side of these nerves, but have no connection with them.

b. *Composition.*— In entire preparations and on cursory examination of sections, the structures may really appear to be vesicles, as Claus said; but closer inspection shows that each organ is composed of two large cells of about equal size, which are so much flattened against each other that they together form an approximately spherical body. Each cell contains a fairly large nucleus (Plate **2**, Fig. 20; Plate **3**, Figs. 31, 33) with chromatin in the form of a network. A comparison of the dimensions of nuclei and cells in the brain proper with those of the organs of Claus, shows that, while the nucleus of a brain cell, on the average, is a little larger than a nucleus of one of the cells in an organ of Claus, the cells of the latter are several times as large as the cells of the brain. The average diameter of nuclei in the cells of the brain is approximately 9.1 μ, while the nuclei of Claus's organ are on the average not over 8.5 μ in diameter. The average diameter of the cells in the brain is 11.7 μ, while the cells in the organs of Claus average 30 μ in diameter. These values have been obtained by careful measurement of cells and nuclei both in whole preparations and in sections cut in three planes.

c. *Similarity in Structure to the Cells of the Median Eye.*— The most striking thing about the cells of the organs of Claus is their resemblance to the cells of the median eye. It may be said that, within certain limits, to be later defined, a cell of an organ of Claus corresponds in every way with a retinal cell of the median eye. Each of the two cells in Claus's organ is provided with a structure which is an exact counterpart of the basal plate in the median eye. It becomes red in Mallory's stain (Plate **5**, Fig. 55, *la. ba.*) and brown or black in vom Rath's mixture (Plate **2**, Figs. 11, 12, *la. ba.*). But in the median eye, a basal plate is shared by several cells, whereas in the organs of Claus each cell has formed its own basal plate, which covers a portion of the periphery of the cell. Sections cut in the three principal planes, and whole preparations, show that the basal plate measured in any direction occupies about one-third of the entire periphery of its cell (cf. Plate **5**, Fig. 55, Plate **2**, Figs. 18 and 20). As in the case of the median eye, the basal plate of the organ of Claus possesses no intrinsic structure. The region of the cells occupied by the basal plates is also, in part, the region in which the cells are in contact. This gives rise to an appearance in darkly stained vom Rath material (shown, for example, in Plate **2**, Fig. 12 and Plate **3**, Fig. 36) which suggests a body suspended in a vesicle, somewhat as an otolith rests in the ear-sac. That this body is really composed of the two basal plates of the apposed cells, which have stained so deeply as to be indistinguishable from each other, is shown when such a preparation is decolorized. Figure 12 (Plate **2**) gives, in a semi-diagrammatic way a rather typical condition that appears in sections of deeply colored vom Rath material; while Figure 11 (Plate **2**) is drawn from the adjoining section of the same organ, after decolorization. Here the line of separation between the two cells and their basal plates is very plain, and from this figure may be learned the reason for the peculiar appearance shown in Plate **2**, Figure 24, or Plate **3**, Figure 36.

The organs of Claus are provided with the interior bodies of Hesse (phaosomes), as are the retinal cells of the median eye. The bodies are similar in all respects in the two structures, and it is therefore unnecessary to enter into a detailed description of their form in the organs of Claus except to state that the band-like appearance of the bodies is very rarely met with. The arrangement of the bodies in the organs of Claus is rather uniformly around or near the periphery of the cell opposite the basal plate. This is seen best in sections (Plate **5**, Fig. 55; Plate **3**, Fig. 36), but also appears in whole preparations; it is difficult, however, to show the condition in proper perspective in a drawing. Figure 33 (Plate **3**) will serve to give some idea of the conditions as seen in an entire preparation. But any description which deals with the arrangement of the interior bodies must be limited to the more general features. A comparison of Figures 33 and 34 (Plate **3**) is instructive as indicating something of the position of the interior bodies in the cells. Figure 33 is a drawing of the organ of Claus shown at the left in Figure 27 and viewed from the dorsal side, while Figure 34 is a drawing of the lateral face of a sagittal section of the same organ. In both, the interior bodies are farther from the basal plates than from the periphery of the cell opposite the plates, but many more are seen in the section (Fig. 34) than were apparent in the whole preparation (Fig. 33), if we limit our consideration to the cell (uppermost in the plate) farthest from the observer in each of the figures. There was really a long peripheral row of rod-like bodies ventral to those actually shown in the anterior cell of Figure 33, but it so happened that in this particular case they were not visible at all.

In general, then, it is a fair statement that the interior bodies of the organs of Claus are peripheral; that is, near that margin of the cell which is farthest from the basal plates. It can scarcely be said that the interior bodies have a definite arrangement or position in regard to the long axis of a cell, as is the case in the median eye; but there is certainly a very definite grouping of these rods in the organs of Claus, and within a group the rods are approximately parallel to each other.

d. *Relation of Nerves to Organs of Claus.*— That the interior bodies in the organs of Claus, as in the median eye, have some functional, if not directly structural, relation to nerve terminations, is at least strongly indicated by such conditions as are shown in Figures 19, 20 (Plate **2**), 26, 29 (Plate **3**). Figure 19 (Plate **2**) is especially good for showing this. It is plain that there is a large bundle of nerve fibrils leaving one of the cells, and that, within the cell, the fibrils are arranged so nearly in the same manner as are the interior bodies, that one can scarcely resist the conclusion that they are interdependent, even if the exact structural relations between the two are not apparent. Likewise in Figure 26, the interior bodies lie directly in the path of the fibrils which go to make up the large nerve (*n.*) leaving the cell, and almost without exception, the long axis of the rod has the same direction as the fibrils. In Figure 51 (Plate **5**) a relatively small nerve may be seen leaving the cell, but here the interior bodies are not visible. They appear in Figure 55, which is drawn from the section adjoining that from which Figure 51 was taken. I have been unable to see that the interior bodies and the nerve fibres are, in any case, structurally connected, but such conditions as are shown in Figures 20 (Plate **2**) and 26 (Plate **3**) offer almost unassailable evidence that their interaction is closely concerned with the sensory function of the organs of Claus.

It is more difficult to ascertain the exact type of nerve-termination in the organs of Claus than in the median eye. But I believe that the type of ending is the same in the two cases, for I have seen in certain preparations unmistakable evidence of nerve fibrils in a cell of the organ of Claus, and they were in every way similar to those of the retinal cells, except that the club-shaped enlargements were *directed toward the basal plate*, instead of away from it. In these cells, also, as in the cells of the median eye, I believe that the denser character of the cytoplasm and the more or less evident striation, and radiate arrangement of particles in the region of the basal plate, is evidence of secretory activity and not of the presence of a "Stiftchensaum."

Notwithstanding the absence of any experimental evidence as to their function, I believe that the structure of the organs of Claus warrants us in regarding them as eyes. The cells of these organs correspond in every essential feature of structure with the retinal cells in the median eyes, since they possess basal plates, interior bodies and nerves. The relation of the latter to the cells is not the same as in the median eye, however. For there we have seen that, whether the "central cell" (pigment cell) or the basal plates are regarded as the pigment-bearing portions of the eye, the nerves leave the retinal cells from their *basal ends*, or that portion of the cell adjoining the pigmented part of the eye, whereas in an organ of Claus, the nerve leaves that portion of the periphery of the cell which is farthest removed from the basal plate (Plate **2**, Figs. 19, 20; Plate **3**, Figs. 26, 29; Plate **5**, Fig. 55). Consequently, if we follow consistently the interpretation hitherto given of the relation between a sensory cell of an optic organ and its nerve fibre, the organs of Claus are to be regarded as *bi-cellular, inverted eyes*. In other words, the ends of the nerve fibrils are directed toward the *bases* of the sensory cells. But in the median eye, the light recipient portions of the optic nerves are directed toward the *outer ends* of the cells.

III. Discussion.

From the previous references to the literature bearing upon the subject, it has appeared that, with the exception of Grenacher ('**79**), all who have studied the median eye of Crustacea have either definitely stated their belief in the inverted character of the retinal cells (Hesse, :**01**, :**02**; Claus, '**91**, Hartog, '**88**; Richard, '**91**), or, taking neutral ground upon this particular point, have felt that a close comparison with the eyes of the flatworms was justifiable (Carrière, '**85**; Lang, '**88**–**94**; Claus, '**63**). A quotation from Hesse (:**02**, p. 630) will put this matter concisely, and since it comes from one whose knowledge of the optic organs in invertebrates is unexcelled, it may be considered as representative. "Die invertirten Pigmentbecherocellen haben eine sehr weite Verbreitung. Alle Sehorgane, die wir bei den Plathelminthen kennen, sind hierher zu zählen: also die Ocellen der Turbellarien, die x-förmigen Augenflecke der Trematodenlarven, und die Ocellen der ausgebildeten ektoparasitischen Trematoden, die Ocellen der Nemertinen und wahrscheinlich auch diejenigen der Rotatorien. Ferner gehören hierher mit grosser Wahrscheinlichkeit die Ocellen der Trochophoralarven und ähnlicher Larvenformen, sicher die Ocellen des Nauplius und die mit ihnen identischen Medianaugen vieler ausgebildeter Crustaceen. . . ." From this point of view it may readily be seen how it is possible for such a statement as the following (Hesse, :**02**, p. 647) to be made: "Bei den Crustaceen sind die Medianaugen nichts Anderes als die übrig gebliebenen Naupliusaugen, die eine Erbschaft von wahrscheinlich plathelminthenartigen Vorfahren darstellen."

The essential point in all these comparisons is the inverted character of the retinal cells of the median eye; it is maintained that the axis cylinders, which are made up by the union of neurofibrillae from within the cells, leave the cells from that part which is directed away from the pigment. But, if the eye of Eucalanus may be taken as the type of the so-called persisting nauplius eye, such a comparison as that instituted by Hesse must fail, as well as the conclusions deduced from it. Hesse himself (:**01**, p. 350) has considered the median eye in Eucalanus as of typical form, as did Grenacher ('**79**, p. 63) and others who followed him (Carrière, '**85**; Lang, '**88–94**); in the absence of trustworthy evidence to the contrary, it seems to me that such a median eye may fairly be taken as representing the general form of the tripartite eye as found among Crustacea. At any rate, the median eye is a more characteristic structure in the Copepoda than in any other group, and, among the Copepoda, the eye of Eucalanus has been more adequately studied than that of any other genus.

But so far, then, as my observations extend, there is no evidence from the manner of innervation of the median eye that it is of the inverted type. For, as Beer (:**01**, p. 12) has said of the uninverted eye "das Licht unter den gewöhnlichen Bedingungen erst die Photirzelle, dann den Opticusabgang trifft."

The median eye of Crustacea has been placed in the same class with those of the flat-worms, and of many annelids, on account of its position as regards the epithelium. Hesse (:**02**, p. 620), in a table giving the results of his investigations, includes the median eye of Crustacea among those whose visual cells are subepithelial. This term is defined (Hesse, :**02**, p. 619) as follows: "Wenn dieser gleiche Vorgang, der eine ursprünglich epitheliale Sehzelle zur intraepithelialen werden lässt, noch weiter fortschreitet, so verlässt die Sehzelle den Bereich des Epithels vollkommen: sie wird zur *subepithelialen Sehzelle*." Claus ('**91**, p. 260) is also of the opinion that the median eye as a whole is separate from the hypodermis. I believe my results prove that only the paired portions of the median eye can be regarded as subepithelial in the sense in which Hesse has used the term. There is very little indication in the adult condition that the lateral eyes are a part of the general ectoderm of the body; consequently they cannot belong to Hesse's class of epithelial eyes, and there is just as little reason for regarding them as "intraepithelial." Hesse's definition of these two sorts of eyes is as follows: Entweder bleiben sie [Sehzellen] in dem Verband des Epithels wie die indifferenten Epithelzellen, d. h. sie reichen mit ihrem distalen Ende ganz bis an die äussere Begrenzung des Epithels . . . . dann sind sie selbst Epithelzellen geblieben, wir bezeichnen sie als *epitheliale Sehzellen*. . . . Wenn dagegen eine Sehzelle mit ihrem distalen Ende nicht bis an die distale Grenze des Epithels reicht, übrigens aber zwischen den Epithelzellen liegt, so ist sie keine eigentliche Epithelzelle mehr, sie ist innerhalb des Epithels gleichsam versenkt, proximad verschoben: wir bezeichnen sie als *intraepitheliale Sehzelle*" (Hesse, :**02**, p. 618, 619).

From these definitions, it is plain that the ventral portion of the median eye is to be regarded as composed of neither *intraepithelial* nor *subepithelial*, but simply of epithelial retinal cells. I have shown that it is continuous at its margins with the undifferentiated ectoderm, and that it is, therefore, merely a thickening in the ectoderm. It evidently is of the first type, which Parker ('**91**, p. 59) has mentioned as characteristic of most of the groups of Crustacea. Moreover, Hesse (:**02**, p. 620) considers that the compound eyes of all arthropods have epithelial visual cells. But it is impossible to regard the sensory cells of the ventral part of the median eye as either intraepithelial or subepithelial.

As regards the cells of the lateral portions of the median eye, they properly belong to those classed as subepithelial by Hesse or to the third of the types enumerated by Parker ('**91**, p. 60), if we are to judge from the conditions in the adult. But Figures 6 (Plate **1**) and 44 (Plate **5**) show, especially at the anterior end, that the membranes around the lateral eyes are a part of the basement membrane of the hypodermis, though it cannot be claimed that the retinal cells and those of the hypodermis are now directly continuous as in the ventral portion. As regards the membrane which surrounds the optic nerve, its relation to the eye as a whole, and especially to the lateral portions of the eye, seems to me to argue for the subepithelial character of the retinal cells, for some of them certainly lie within the membrane, which is plainly subepithelial in position.

If, then, we attempt to place the median eye (as known in Eucalanus) in the morphological classification proposed by Hesse (:**02**, p. 620), it is necessary to consider that the unpaired portion occupies one position in the system and that the paired portions occupy another. The ventral ocellus is epithelial; the lateral ocelli are subepithelial. And none of the cells of any part of the eye can be considered as inverted, even through reversion, as Hesse (:**02**, p. 620 and 631 *ff.*) has maintained is the case among certain flatworms and the leeches.

The relations of the cells of the eye to the hypodermis and to the nerves are important in considering the phylogenetic position of the median eye. Most, if not all, investigators have been inclined to refer the median eye to the pigment-spots on the apical plate of annelid larvae, and through these to the flatworms. Claus ('**91**, p. 260) states that the three optic cups of the median eye of Crustacea "phylogenetisch vielleicht mit den Punktaugen an der Scheitelplatte von Anneliden-larven in Beziehung zu bringen sind." And Hesse (:**02**, p. 644) draws a similar comparison; the inverted pigment-cup type of ocellus is found in all the flatworms or is derived therefrom by reversion. "In die Verwandtschaft der Plathelminthen gehört zweifellos die Trochophoralarve, der wir wohl die Naupliuslarve zugesellen dürfen. Von diesen Larven ist vielleicht diese Form der Sehorgane auf die fertigen Thiere übergegangen: so finden wir sie bei niederen Anneliden, und zwar meist dem Gehirn anliegend, wie sie bei der Larve in der Scheitelplatte liegen, und bei den Medianaugen der Crustaceen ist es ja sicher, dass sie die persistierenden Naupliusaugen sind." Hesse (:**02**, p. 647) also states that the median eye in Crustacea is nothing more than a structure inherited from ancestors probably like the flatworms. And both Lang ('**88–94**, p. 421) and Korschelt und Heider ('**90**, p. 386) consider that the nauplius is referable to a trochophore larva. Crustacean characters are concealed (zurückverlegen) in the nauplius, though the median eye is not mentioned as one of these characters. Lang is strongly of opinion that the nauplius larva does not represent the aucrestral crustacean form. The latter is, in his opinion, to be sought among the worms, and the nauplius is to be regarded as a typical crustacean larva, which possesses many primitive characters of Crustacea. Finally, Zograf (:**04**), according to his reviewers, looks upon the unpaired eye as an organ that occurred in the primitive Crustacea, since it is present in the larvae or embryos of all Crustacea, and even persists in certain of the higher forms. Claus ('**61**) proved its presence in the larva of malacostraca. (See also Hartog, '**88**, and Balfour, '**80**, p. 417 ff.)

These references show the manifest tendency of investigators to consider the "nauplius eye" as a primitive one (Claus, '**63**, p. 44), and to base upon this, as well as other characters, the argument for the relationship between worm-like forms and the Crustacea.

I believe that the facts I have brought forward show that, as far as regards the median eye of Eucalanus, which has been held to be a typical nauplius eye, a comparison of it with any of the typical eyes found among flatworms or the lower annelids is not justified. Consequently, I do not feel that the conclusion of Hesse (:02, p. 644) already quoted is warranted at present, especially in view of the complete absence of embryological facts to support his contention.

But that there are light-recipient organs among Crustacea which are in every way justly comparable to those of the lower annelids and the flatworms, is shown, I believe, by the facts already brought forward concerning the structure of the organs of Claus, and by comparative evidence derived from the group of worms.

Von Graff ('82, p. 115) has shown that in the rhabdocoele flatworms, the pigmented and lens-bearing eyes are situated directly upon the brain, and Carrière ('85, p. 25) states that eyes to the number of two or four occur inside the brain.

The investigations of Hesse ('96, p. 404) led him to conclude that the structures which function as eyes in the Lumbricidae are located in the brain as well as in the epidermis. Among the Capitellidae, also, Hesse ('99) has shown that the cup eyes are found both in the brain and in the epidermis, and this holds for many other forms of the limicolous annelids. The position of the organs of Claus, however, has a deeper significance, if we look upon them as *subepithelial.* According to Hesse (:02, p. 620) the eyes of Plathelminthes, the cup eyes of Capitellidae (in part), and of many polychaete annelids are subepithelial, as well as the median eye in Crustacea. But it appears that only the lateral portions of the nauplius eye can be regarded as subepithelial, while the entire organ of Claus must be so regarded. Since the organs of Claus lie entirely within the brain, which has lost all connection with the ectoderm (being separated from it by a thick membrane), they are neither epithelial nor intra-epithelial as these terms have been defined.

In *position,* then, the organs of Claus may be considered as homologous with the simpler eyes found among the flatworms and certain groups of the annelids. Hesse ('99, p. 477) is of opinion that the "Becherauge" is of the same form as that generally distributed through the Plathelminthes, the type being found in Planaria torva; also (p. 483) that the goblet eyes in the lower annelids are essentially like those of the planarians.

It is my belief that the facts previously presented warrant us in extending this statement of Hesse to the organs of Claus found in the brain of Eucalanus. I have shown that the axis cylinders which pass from the visual cells of the median eye to the brain, leave the *basal* *ends* of the cells. And it should be pointed out again that, whether we consider the "central cell" or the "basal plates" as the pigmented portion of the eye, the truth of the previous statement is not affected. In the ventral ocellus of the median eye the basal plates lie beneath the *basement membrane* of the hypodermis from which the retina is formed. Moreover, it is merely consistent, and warranted by the facts of their structure, to consider that the basal plates of the lateral eyes are at the *basal* ends of the cells. If the central cell contains the pigment of the eye, as I believe is the case, the "basal" ends of the retinal cells must also be considered their "inner" ends, since in that case they are directed toward the pigment of the eye.

In the organs of Claus, on the contrary, the nerve leaves the cell from a portion of the periphery *opposite* to the "basal plate." I can see no reason why the structures in the organs of Claus which I have termed "basal plates" should not be considered the homologues of the similar structures in the lateral eyes. Their appearance and staining reactions in the two places are the same in every way. Consequently, it is only reasonable to look upon that portion of a cell of an organ of Claus which bears the plate as the basal or inner portion.

If this interpretation is correct, we are forced to regard the organs of Claus as *inverted*, and as *eyes* because of the complete correspondence in essential features with the cells of the median, nauplius eye. The manner of innervation of these specialized cells in the brain is in accord with the facts recounted by Hesse ('**98**, p. 483), who says with regard to the eyes in the brain of annelids, "An einzelnen Schnitten konnte ich auch beobachten, wie der ausserhalb des Pigmentbechers gelegene Theil der Sehzelle sich zu einer Nervenfaser auszog."

It would seem at first thought that a complete correlation between the organs of Claus and the inverted eyes of worms would force the interpretation that the "basal plates" are pigment-bearing structures. But this is not necessarily the case, for it is the *position* of the plate with regard to the place of exit of the nerve of a cell, which is, as it appears to me, of importance, and not whether the plate may be pigmented or not. If we interpret as broadly as possible the facts of the structure of the median eye, we are compelled to admit that the pigment, wherever it is located, must occupy the same relative position with regard to the point of exit of the optic nerves as the basal plates.

Again, the probability that the basal plates of the organs of Claus are not structurally comparable to the pigment-cells of the eye of a planarian, does not in the least militate against looking upon the organs of Claus as having the function of eyes. Beer (:**01**, p. 13) and Hesse (:**02**, p. 610) have recently pointed out that pigment need not necessarily be present in order that light may stimulate an organ. And Helmholtz ('**56**) long ago expressed the same view: "Doch wissen wir durchaus nicht, ob alle pigmentirten sogenannten Augenpunkte der niederen Thierformen wirklich zur Lichtempfindung dienen. Andererseits müssen wir aus der Empfindlichkeit, welche niedere Thiere ohne Augenpunkte für das Licht zeigen, schliessen, dass auch lichtempfindende Nerven in durchsichtigen Thieren ohne Pigment vorkommen, die nur der Beobachter in keiner weise als solche erkennen kann."

Moreover, the great variability in the position of the pigment in eyes that must be regarded as homologous, as Hesse (:**02**, p. 613) has shown, is also against the view that a visual cell, as such, must be pigmented.

It is unnecessary, and probably unwise, to speculate at length as to the manner in which the median eye in Eucalanus, or other animals with a similar organ, has been formed, because embryological evidence along that line is entirely lacking. I have already pointed out that the conditions in Eucalanus favor the view that the lateral eyes are subepithelial, having virtually lost their original position in the ectoderm. The simplest explanation for the adult condition seems to me to be that each of the vesicles forming the lateral eyes has become, by a simple revolution through 90° or more in opposite directions, so oriented that the ends of the cells which were at first ventral are now directed dorso-laterad. Thus in the paired eyes the ends of the cells which were originally covered by the basement membrane of the hypodermis have become, at least partly, reversed and are now directed medio-ventrad. This condition may be imagined as resulting from the rolling in of the lateral portions of a once-continuous sensitive area embracing all three of the components of the median eye, until the lateral margins of the area nearly meet in the median plane of the body, the middle ocellus undergoing in this revolution no change of position.

But such alterations in the position of the eye as a whole as may be assumed to have taken place, have evidently not resulted in the formation of structurally *inverted* cells in the lateral eyes. Claus ('**91**, p. 260) has assumed that as the eyes separated from the hypodermis there occurred "eine convergent nach einen Punkte gerichtete Drehung . . . . um eine Erklärung für das Zusammenstossen ihrer convexen Flächen und den Eintritt der Nerven von der Aussenseite in die Retina zu gewinnen." Through this turning the "ursprünglich abwärts gerichteten Eintrittsstellen der Nerven in die Retina" came to assume a more lateral and superficial position, and so the more or less distinctly inverse form of the eyes has arisen. We must admit that the parts of the eye do not begin their development in the positions which they come to occupy in the adult; but in Eucalanus the change has not brought about inversion of the retinal cells. Hesse (':**01**, p. 353) takes a view similar to that of Claus. He is of opinion that the inversion of the median eye of Crustacea occurs because visual cells, which have migrated out of the epidermis, have become oriented, for physiological reasons, with the sensitive portions directed toward the pigment; later the cup about the cells is formed by the incurving of the pigmented shell. It is probable that the relative positions of the ventral and lateral ocelli are to be interpreted as indicating that the lateral eyes in such forms as the Pontellidae are originally portions of the median eye. Leuckart ('**59**, p. 260) seems to be of the opinion that the eyes of Anomalocera, which are provided with lenses, are independent of the median eye, in the sense that the latter is the equivalent of the eye of Cyclops. Claus ('**59**) at first accepted this view, but later ('**63**, p. 46) was led to believe that the lateral eyes of Pontellidae might be considered as originally belonging to the median eye. In a subsequent paper (Claus, '**91**, p. 247), however, he again adopted the interpretation of Leuckart and states (p. 250) that the dorsal eyes of the Pontellidae must be very different from the median eyes, and are to be homologized with the compound, facetted, eyes of Arthropoda.

Parker ('**91**) has shown that the lens eyes of Pontella are entirely separated from the body ectoderm, while the compound eyes of the Decapoda are continuous with the hypodermis. Those of the Cladocera and Branchiopodidae are of an intermediate type. Hesse ('**02**, has adopted a similar classification as regards the higher Crustacea and Arthropoda in general. With this I agree, and consequently, I think we cannot adopt the view of Claus ('**91**, p. 250), that "Wir haben also die interessante Thatsache zu constatiren, dass auch unter den Copepoden [Pontelliden] das bei den Phyllopoden und auch Cirripedien schon so hoch entwickelte zusammengestzte Augenpaar vertoeten ist." Whether the lens eyes of Pontella are compound or not, they must be looked upon as of an entirely different type from those of the Decopoda or Phyllopoda and Cladocera.

But as regards Eucalanus, the conclusion seems to me to be warranted that the lateral ocelli are subepidermal and, in that particular, of the same type as the lateral eyes of Pontella. The axes of the cups are practically the same in the two cases. Taking into consideration the fact of the position of the retinal cells with reference to the hypodermis, the lateral ocelli of the median eye of Eucalanus may be homologized with the lateral eyes of the Pontellidae. The latter are, it is true, more specialized, but were originally portions of the "nauplius" eye. The conditions in Eucalanus show, I believe, the first steps in a process by which the lateral ocelli (as represented in Pontella) have left the ventral ectoderm of the body and the ventral ocellus of the tripartite eye, and have finally become both subepithelial and dorsal in position. The ventral eye of such forms as Pontella (in the adult) is the remaining (median) portion of the median eye, and not the homologue of the entire tripartite median eye. Claus ('**91**, p. 248) based his conclusions upon the structure of the ventral eye of Pontella. He found that it consists of a middle and two lateral portions and therefore, in his opinion, corresponds to the median eye with its three pigment cups and retinas.

Zograf (:**04**), with whose work I am acquainted only from reviews, assigns a later origin to the paired ocelli of the median eye than to the ventral part, and considers that the three have become united only secondarily.

In all of his papers, Hesse has laid much stress upon the character of the nerve endings in the visual cells. In his concluding discussion (:**02**, p. 598) he distinguishes two sorts of visual cells on the basis of the recipient parts: those with free endings of neurofibrillae, and those with "*Phaosomes*," the latter being by far the less numerous. I have already mentioned the extent to which, in Hesse's opinion, the free nerve endings occur in the form of a "Stiftchensaum." He (Hesse, :**02**, p. 608) applies the term phaosome to intracellular bodies in the visual cells of Naidae and Lumbricidae. "Die Ausdrücke, welche bisher für diese und ähnliche Gebilde gebraucht sind, wie Vacuolen, Binnenkörper, Glaskörper, sind zu unbestimmt, oder auch für andere, durchaus verschiedene Objekte in Gebrauch, so dass ich sie lieber nicht gewählt habe." If I understand his idea correctly, he considers it possible that free nerve endings are found in cells which possess phaosomes. "Jedenfalls ist zu vermuthen, dass auch in den Sehzellen mit Phaosomen, wie in allen Zellen des Nervensystems, Neurofibrillen vorhanden sind." "So lange diese in ihrem Verhalten zu den Phaosomen nicht nachgewiesen sind, muss jede Vermuthnug, ja sogar die Annahme, dass die Phaosomenzellen von denen mit freien Neurofibrillenenden grundsätzlich verschieden seien, als provisorisch erscheinen..... "Die Verbreitung der

Sehorgane mit Phaosomen ist also eine sehr geringe, so gering, dass wir sagen können in fast allen Sehorganen würden die Lichtrecipirenden Endeinrichtungen der Sehzellen durch freie Neurofibrillenenden gebildet" (Hesse, :02, pp. 609, 610). This author also states (pp. 608, 609), that the most evident ground for assuming that the phaosomes play an essential part in the visual cells, lies in their constant occurrence in those cells only which may rightly be considered as visual cells. Further, as the first of the above quotations shows, he thinks, it may be assumed that neurofibrillae are present in the visual cells with phaosomes as they are in all other cells of the nervous system.

I have given my reasons for concluding that free nerve endings occur in the sensory cells of the median eye of Eucalanus, but I do not agree with Hesse (:01) in his statement that the endings are of the widespread type which he calls the "Stiftchensaum." It would be presumptuous to doubt the occurrence of this structure (Stiftchensaum) in the other forms in which he states that it is found, but one may infer from my results that a possible reason for the non-occurrence of a "Stiftchensaum" in Eucalanus is that Phaosomes are present. At any rate, I can see no reason for not regarding the "interior bodies" as phaosomes, and Hesse himself (:02, p. 609) states that the intracellular, bandlike bodies in the visual cells of several Calanidae are comparable to phaosomes.

There is also reason for believing that the phaosomes are intimately connected with the transformation of the energy of light into that of the nervous impluse. The club-shaped ends of the neurofibrils have been seen in some cases to lie in contact with the phaosomes, and the latter have in general a striking arrangement within the cells, more or less conformable to the courses of the nerves. We may infer, then, that there is more reason for assigning to the phaosomes an important rôle in the visual cells, than simply that they constantly occur there. It seems not improbable that the phaosomes are, in a way, comparable to the rhabdome of higher crustacea, for it is well known that in the decapods the optic nerve fibres terminate in the rhabdome (Parker, '95, p. 20). Hesse (:01, p. 435) regards the rhabdome of Astacus as composed of the seven modified "Stiftchensäumen" of the seven retinular cells, and Parker ('91, p. 81) is of the opinion that the "rod-like bodies" in the retina of Pontella probably represent rhabdomeres.

It would be interesting if it could be shown that the median eye in Copepoda is a forerunner of the compound eyes. It is not absurd to look upon the epithelial character of the ventral portion of the median eye as pointing in this direction. The possession of phaosomes, or rhabdome-like bodies, and the manner of innervation (the cells are not inverted) are also evidence along the same line. But the facts at hand do not warrant an extended discussion of this matter, nor at the present time more than a reference to the possibility of such a relation.

The more or less prevalent opinions as to the relation existing between the median eyes of the Crustacea and the eyes of lower forms, such as flatworms, have been referred to previously. And it has been shown, that, so far as the conditions in Eucalanus may be taken as indicative, the reference of the median eye to those of flatworms, for example, is not warranted. On the other hand, the organs of Claus are believed to be in *every essential point* comparable to the inverted eyes of other invertebrates, more particularly to those of the annelids; yet they exhibit a close similarity in the structure of their cells to the median eye.

It cannot be maintained that a single character, such as the structure of visual cells, may in itself be reasonably regarded as showing racial affinity. Yet the facts that the cells of the organs of Claus in Eucalanus are probably eyes and if so are of the *inverted, subepithelial* type, seem to me to be evidence, along unsuspected lines, of the derivation of crustacean from annelidan stock, which as already mentioned, has been rather generally, and on other grounds, looked upon as probable (Lang, **'88–94**, p. 419). The character of the nerve-terminations as such in the visual cells does not seem to me to be of as fundamental importance as the other matters I have referred to. I think it probable that even in the organs of Claus the neurofibrillae are not in the form of a "Stiftchensaum," but the observations are not based upon as clear conditions as in the cells of the median eye. Whether a visual cell is inverted or not has always been regarded as of wide significance, and when cells are met with that possess this character, such as those of the organ of Claus, the meaning is worthy of consideration. As regards the derivation of the visual organs of one group of animals from those of another, Carrière (**'85**, p. 201) is of opinion that all known facts are against it, and that, on the contrary, the visual organs are "convergent structures." Hesse (**:02**, p. 643) says, with regard to Carrière's idea: "Wir können diesen Aüsserungen nach dem Vorhergehenden unmöglich beistimmen"; further (Hesse, **:02**, p. 646) "Dass aber gegenüber anderen Organen die Sehorgane eine Ausnahmestellung einnähmen, derart, dass sie weinger von einer Gruppe zur anderen vererbt würden (Carrière), muss auf Grund der Thatsachen durchaus bestritten werden." But at the same time, he justly states that the relationships of various groups can be determined neither by the structure of one organ nor of one system of organs; the entire organization must be considered.

With these considerations in mind, it may fairly be concluded that the occurrence of inverted "Pigmentbecherocellen" among Copepoda, points strongly toward the relationship of this group with the polychaete annelids, particularly if the entire structure of the organisms is taken into consideration. On the other hand, the median eye of the calanoid Copepoda can not from its *structure* be regarded as "eine Erbschaft von wahrscheineich plathelminthenartigen Vorfahren," even though it may represent the persistent nauplius eye.

IV. Summary.

1. The median eye of Eucalanus is of the well-known tripartite type. Each lateral ocellus consists of two *basal plates,* and of nine *retinal cells.* The ventral ocellus of the eye contains ten cells, and is provided with a single basal plate similar to those of the lateral portions of the eye.

2. The basal plates are products of the retinal cells, and probably do not contain the pigment of the eye. This is believed to lie in a *central cell,* upon or in which the three divisions of the eye rest. The *tapetum* lies upon the peripheral margins of the central cell.

3. The retinal cells are provided, in their cytoplasm, with "interior bodies" or *phaosomes.* These have generally a flattened rod-like form and are arranged in such a way that when sectioned the long axis of their section corresponds with the long axis of the section of the cell, whatever the plane of section may be.

4. The axis cylinders of the optic nerves leave the retinal cells at the *basal* or *deep ends* (those adjoining the pigment cell), and pass through, or to one side of, the basal plates to enter the central cell. The individual fibres traverse the central cell toward the brain.

5. There are twenty-eight fibres in the optic nerve. The number corresponds exactly with the total number of retinal cells in the entire eye, and it is therefore highly probable that one fibre comes from each cell. The fibres may be traced in their individuality some distance into the brain.

6. The terminations of the nerves in the sensory cells are not in the form of a "Stiftchensaum." The neurofibrillae are rather irregu-

lar and somewhat beaded and branched; each terminates in a club-shaped enlargement.

7. Consequently the character of the nerve-ending cannot be regarded as similar to that in the visual cells of worms, as Hesse has maintained.

8. The cells of the median eye are not of the inverted type commonly found among flat-worms and polychaetes. Therefore the median eye is not to be regarded on this character as a structure inherited from worm-like ancestors.

9. The "interior bodies" and neurofibrillae seem to be structurally continuous. Their functional interrelationship is therefore probable.

10. The ventral division of the median eye is simply a thickening of the hypodermis of the body, and has retained, in the adult, its original position. The lateral divisions of the eye have lost all except a very slight connection with the hypodermis. The ventral division is in position epithelial; the lateral divisions are, in effect, subepithelial.

11. These relations are interpreted as evidence that the lateral ocelli of the median eye of Eucalanus are homologous with the lens eyes of the Pontellidae. The ventral ocellus in Eucalanus corresponds to the ventral eye of Pontella.

12. The organ of Claus is to be regarded as a *bicellular, inverted eye*. These organs are located symmetrically in the brain.

13. Each cell of an organ of Claus possesses a basal plate and "interior bodies." These structures are in every respect similar to those found in the cells of the median eye.

14. The nerves from the organs of Claus do not pass through the basal plates, but leave the periphery of the cell at a point which is *opposite* to the basal plate. In comparison with the retinal cells, these are consequently inverted.

15. In position the organs of Claus are subepithelial, and since they lie in the brain as well, they are strictly comparable to the inverted pigmented ocelli of certain worms.

16. Consequently, it is believed that if a relationship between the Copepoda and the groups of the worms is to be sought on the basis of the structure of the optic organs, it must be through the organs of Claus and not through the "median eye." Heretofore the median eye has generally been regarded as inverted, and on this character likened to the eyes of flatworms, which present that condition; but the median eye of Eucalanus gives no support to that view.

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Explanation of Plates.

All the figures were drawn with the aid of a camera lucida. In general, dark tones are to be interpreted as meaning that the structures so represented are at a higher focus in the preparation than those shown in a lighter tint. This is particularly true of the "interior bodies" and nuclei; simple lines need not necessarily be so looked upon. All figures are of Eucalanus elongatus Dana.

ABBREVIATIONS.

a.	Anterior.
cb.	Brain.
cl. c.	Central (pigment) cell.
cl. cb.	Cell of brain.
d.	Dorsal.
dx.	Right.
fbr.	Fibre (or fibres) of optic nerve.
h'drm.	Hypodermis.
la. ba.	Basal plate.
mb. ba.	Basement membrane of hypodermis.
mb. pi'n.	Membrane surrounding optic nerve.
n.	Nerve from organ of Claus.
n. at.	Antennal nerve.
n. f.	Frontal nerve.
n'fbrl.	Neurofibrillae.
nl.	Nucleus of retinal cell.
nl′.	Nucleus of cell of organ of Claus.
nl″.	Nucleus of central (pigment) cell.
nl.h'drm.	Nucleus of hypodermal cell.
n. opt.	Optic nerve.
n'pil.	Neuropil entered by optic nerve.
o. Claus.	Organ of Claus.
ocl. dx.	Right ocellus of median eye.
ocl. m.	Median (ventral) ocellus or median eye.
ocl. s.	Left ocellus of median eye.
p.	Posterior.
par. cp.	Body wall.
pha'so.	Phaosomes or "Interior bodies."
s.	Left.
tap.	Tapetum.

PLATE 1.

Fig. 1. The median eye seen from the dorsal side. Entire preparation, fixed in vom Rath's fluid 12 hours. The optic nerve is shown, also the frontal nerves. There is some indication of the typical arrangement of the "interior bodies" or "phaosomes," in the retinal cells. The nine nuclei in each of the lateral ocelli are shown. × 270.

Fig. 2. Frontal section through the lateral ocelli, seen from the dorsal side. Zenker; iron-haematoxylin; orange G. Optic nerve fibres are seen converging to form the optic nerve. The nucleus of the central or pigment cell and the tapetum, which covers its outer faces, are both shown. × 665. (Cf. Plate **6**, Fig. 59.)

Fig. 3. A parasagittal section of the eye, seen from the left side. Mallory's connective-tissue stain. The tapetum lies on the margins of the central cell, dorsal to the ventral ocellus, and ventral to the lateral ocellus. The basal plates are shown in part on each of the ocelli and it can be seen that the ventral ocellus is merely a thickening in the hypodermis. Five of the optic nerve fibres (*fbr.*) cut across are shown in the central cell. × 665.

Fig. 4. Sagittal section of the brain, to show optic nerve entering at its anterior end, and the optic neuropil. Mallory's connective-tissue stain. × 270. Compare with Figures 35 (Plate **3**), 42 and 43 (Plate **4**).

Fig. 5. Frontal section similar to that shown in figure 2. Vom Rath's fluid 24 hours. The basal plates and tapetum have stained intensely black. The arrangement of the interior bodies (*pha'so.*) in the cells is typical. × 665.

Fig. 6. Sagittal section of entire eye seen from the right side. Zenker; iron-haematoxylin. The ventral ocellus lies in the hypodermis, and the basal plates are shown on its dorsal surface. The optic nerve is made up from three sets of fibres, which lie in the plane of the section; the fibres from the ventral ocellus pass through, and posterior to, the basal plate. × 665. Compare with Figure 51 (Plate **5**).

Fig. 7. Transverse section through the middle of the eye. Vom Rath's fluid 18 hours. Intended to show principally the cells of the eye and the arrangement of the interior bodies with reference to the long axes of the cells. The finely striated character of the basal parts of the retinal cells is shown. × 665. (Cf. Plate **6**, Fig. 58.)

Fig. 8. Frontal section of ventral ocellus. Zenker; iron-haematoxylin; orange G. The ten retinal cells and their nuclei are shown. The arrangement of the interior bodies in the cells is typical. Rod- or spindle-shaped bodies are shown, as well as the ribbon-like forms. × 665.

FIG. 9. Cross-section similar to the one shown in Figure 7. Vom Rath's fluid 24 hours. The fine striation of the inner ends of the retinal cells is shown. The interior bodies have a rather definite arrangement, but they are of unusual shapes. × 485.

FIG. 10. Entire eye seen from dorsal side. Vom Rath's fluid following formalin. The preparation has been distorted by pressure. Ten nuclei appear in the ventral ocellus, and nine in the right lateral ocellus. × 485.

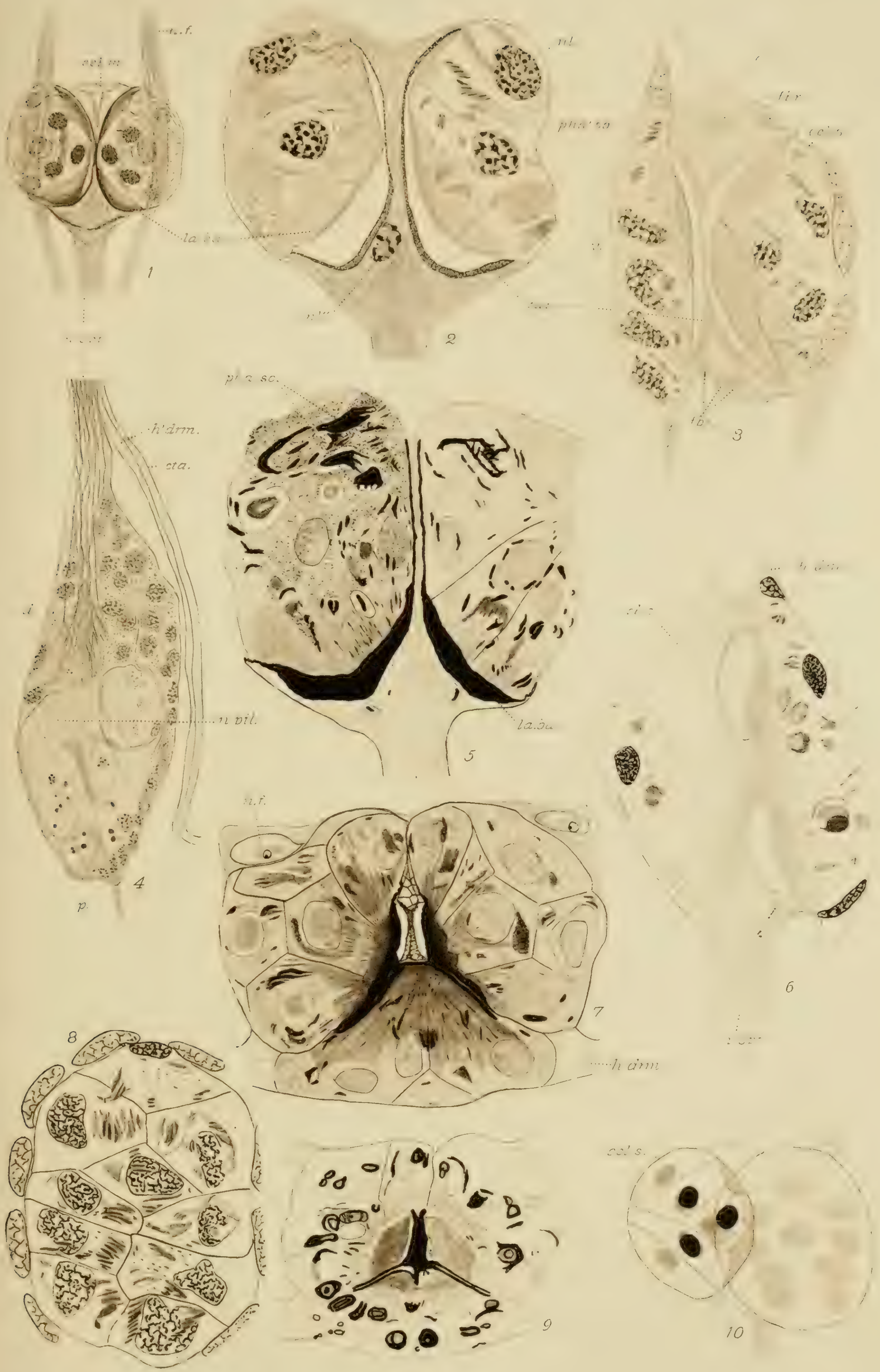
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PLATE 2.

Fig. 11. Oblique frontal section of the left organ of Claus, seen from dorsal side. Vom Rath's fluid 18 hours; decolorized in hydrogen peroxide. The two cells composing the organ are shown, and their basal plates; the interior bodies lie near the periphery of the cells. The striate appearance of the cell contents should be noted. × 665.

Fig. 12. Dorsal aspect of a frontal section of the same organ of Claus as is shown in Figure 11, but the succeeding section and before decolorization. It shows the blackening effect of vom Rath's fluid on the basal plates, and that the basal plates of two cells look like a single body suspended in a vesicle. × 665.

Fig. 13. An organ of Claus seen from the ventral side. Vom Rath's fluid 18 hours; pyroligneous acid 4½ hours, decolorized in hydrogen peroxide. The ventral cell with strong outline has been drawn in detail; the dorsal cell is outlined very faintly. × 665.

Fig. 14. A drawing of the organ of Claus from which Figures 13 and 15 were also drawn. In Figure 14 the focus was fixed at about the middle of the thickness of the organ, to show how the combination of the basal plates of the two cells gives rise to the appearance of a solid body suspended in a vesicle. × 665.

Fig. 15. The dorsal cell of the organ of Claus, the remaining portions of which are shown in Figures 13 and 14.

Fig. 16. Anterior aspect of a cross section of eye. Zenker; iron-haematoxylin; orange G. The difference in appearance between the nuclei of retinal cells and of hypodermal cells is shown, as well as the relation of the tapetum and basal plates. × 665.

Fig. 17. Very oblique cross section (partly *frontal*) of eye. Vom Rath's fluid 24 hours. Particularly favorable for showing a typical arrangement of the interior bodies, and the striation of the inner ends of the cells. The arrangement of the interior bodies corresponds with the direction of the fibres in the optic nerve outside the eye. × 665.

Fig. 18. A cross section through the anterior part of the brain seen from in front. (Compare Plate 6, Figure 61.) Vom Rath's fluid 24 hours; decolorized with hydrogen peroxide. The left organ of Claus is in contact with the nerve of the first antenna; the brain, nerve, and organ of Claus rest upon the hyodermis. The striate appearance, due to the arrangement of granules in the cells of the organ of Claus, is shown. × 665.

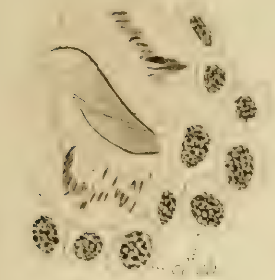
Figs. 19, 20. Frontal sections of an organ of Claus, seen from the dorsal side. Vom Rath's fluid 18 hours; decolorized in hydrogen peroxide. The nerve is clearly shown leaving the cell from a point on the periphery opposite to the basal plates. The interior bodies are arranged in typical fashion. Figure 19 is ventral to Figure 20. × 665.

Fig. 21. The entire eye seen from the ventral side. Vom Rath's fluid 18 hours; pyroligneous acid 4½ hours; decolorized in hydrogen peroxide. The ventral ocellus, the boundaries between its cells, and the nuclei are shown by sharp dark outlines. The preparation is slightly distorted by pressure. The typical and general arrangement of the interior bodies as described in the text is well shown. ×485.

Fig. 22. The eye, entire, from the dorsal side. Vom Rath's fluid following formalin. Ten nuclei are shown in the ventral ocellus, nine in each of the lateral ocelli. The nuclei which are shown in black lie at the highest focus, those with palest tint at deepest focus; the nuclei of the ventral ocellus are merely outlined. × 560.

Fig. 23. Cross section of the eye, seen from in front. Zenker's fluid; iron-haemotoxylin. Shows the striation of the basal portion of the retinal cells, also the interior bodies, the basal plates and the pigment cell. × 665.

Fig. 24. Anterior part of brain seen from dorsal side. Vom Rath's fluid one hour; pyroligneous acid one hour; decolorized in hydrogen peroxide. The organs of Claus are shown in position at the front lateral margin of the brain, near the antennal nerve. The optic nerve is median. × 270.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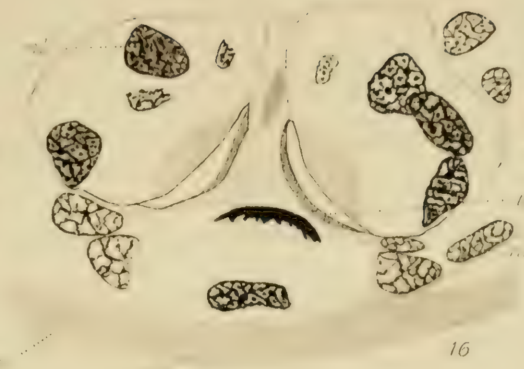
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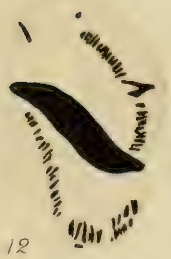
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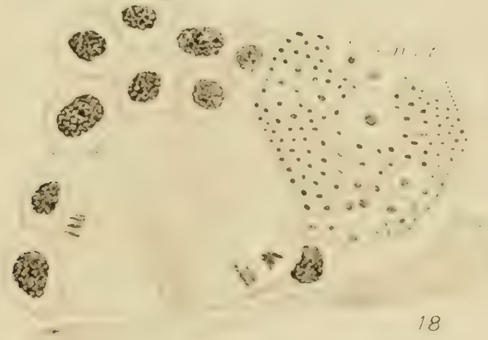
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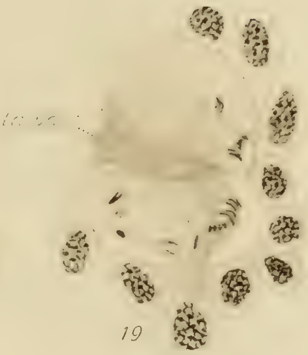
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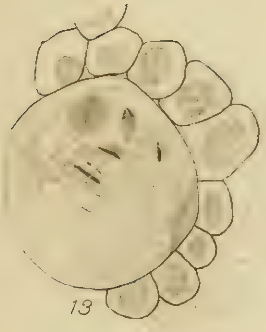
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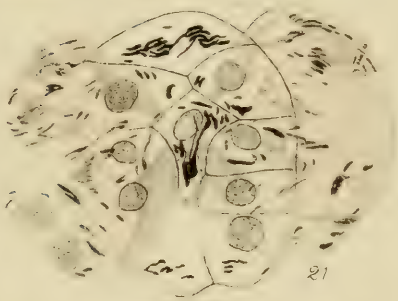
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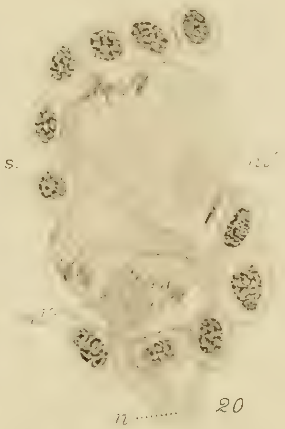

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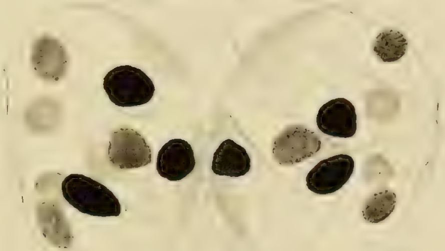
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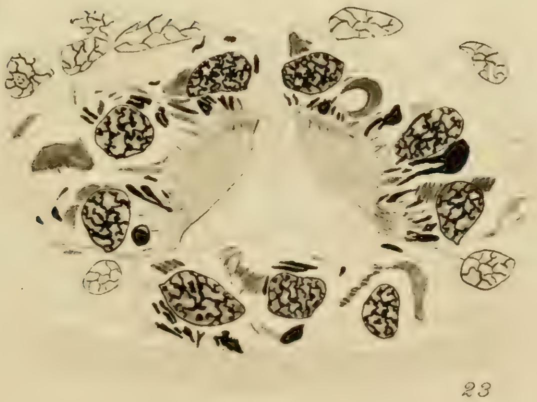
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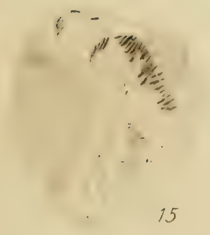
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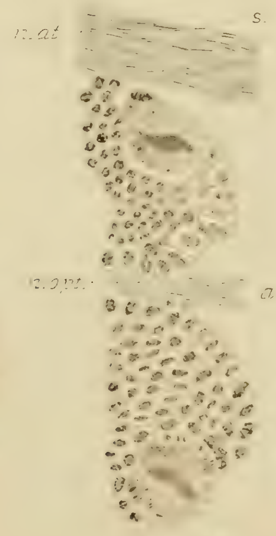

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PLATE 3.

FIG. 25. Dorsal aspect of part of a frontal section of the brain. Vom Rath's fluid 18 hours, decolorized in hydrogen peroxide. It shows one cell of the right organ of Claus and its nerve in relation to the neuropil mass of the brain. The section is the most ventral of three which show the organ of Claus. The basal plate was removed with the two more dorsal sections, so it is evident that the nerve leaves the cell on the side opposite to that of the basal plate. × 405.

FIG. 26. The cell of the organ of Claus shown in Figure 25, enlarged to 665 diameters, showing the course of the fibrillae and the arrangement of phaosomes.

FIG. 27. Anterior part of brain, organs of Claus and parts of optic nerve, frontal nerves, and antennal nerves. Entire preparation seen from dorsal side. Vom Rath's fluid 18 hours, pyroligneous acid 4½ hours; decolorized. × 305.

FIG. 28. Right face of a sagittal section of an organ of Claus. Vom Rath's fluid 18 hours; decolorized. The general form of the two basal plates as seen in this section is the same as in cross sections (Plate **2**, Fig. 18) or in frontal sections (Plate **2**, Fig. 11). The interior bodies are rod-like and lie around the periphery of the cell. × 665.

FIG. 29. The most dorsal of the frontal sections of the organ of Claus in the series of which Figure 26 is the most ventral. The intermediate bodies, part of the basal plate, and nerve are shown. The nerve leaves the cell at a point opposite the basal plate. × 665.

FIG. 30. The middle section in the series shown in Figures 29, 30 and 26. The interior bodies are few compared with those found in the other two sections, and the extent of the basal plates is greater. × 665.

FIG. 31. Ventral view of entire organ of Claus. Vom Rath's fluid 18 hours, pyroligneous acid 4½ hours, decolorized. The deep (dorsal) cell has been drawn in detail (outline pale); the ventral cell is drawn with a sharp outline, but besides the outline only the nucleus and some phaosomes are reproduced. The basal plates are not shown in either cell. × 665.

FIG. 32. Anterior face of a cross-section of a part of the brain, the right organ of Claus and right antennal nerve. The organ of Claus, it will be observed, is on the opposite side of the brain from that shown in Plate **2**, Figure 18. The difference between the two cases in the position of the line of contact of the two cells should be noted. × 665.

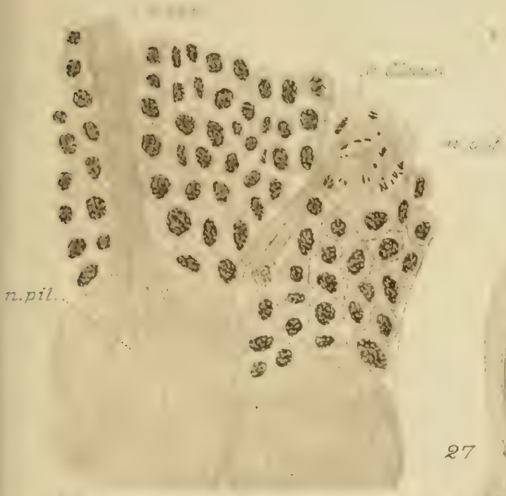
FIG. 33. Enlarged drawing of the left organ of Claus shown in Figure 27. The nature of the basal plates is shown to some extent. × 665.

FIG. 34. Sagittal section of the left organ of Claus shown in Figure 27. The plane of section is indicated by the line of α, β in Figure 33. The depth of the structure is revealed by Figure 34, but was not brought out in Figure 33. × 665.

FIG. 35. Semi-diagrammatic drawing of frontal section of brain and part of the circumoesophageal commissures. Mallory's connective-tissue stain. To show relations within the brain of organs of Claus, nerves and neuropil masses, as well as nerve fibres. This section should be compared with the sagittal section shown in Plate 1, Figure 4. × 270.

FIG. 36. Cross-section of an organ of Claus. Vom Rath's fluid 18 hours. Inserted to show shape of basal plates in deeply blackened preparations, and the mutual arrangement of the two cells, and their granules and interior bodies. × 665.

FIG. 37. Frontal section of organ of Claus seen from dorsal side. Vom Rath's fluid 18 hours; decolorized. It shows the line of contact between the cells of the organ, and the arrangement of granules and of interior bodies. × 665.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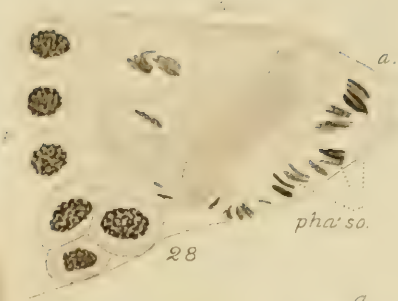

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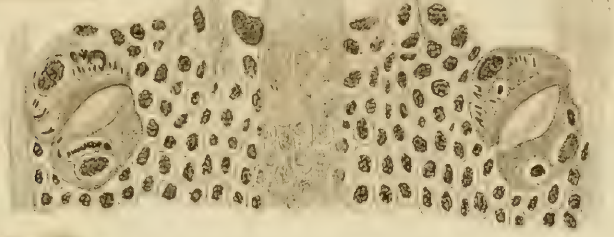

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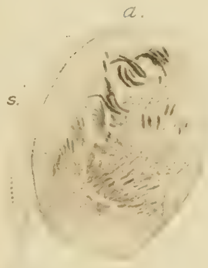

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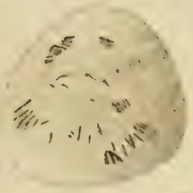

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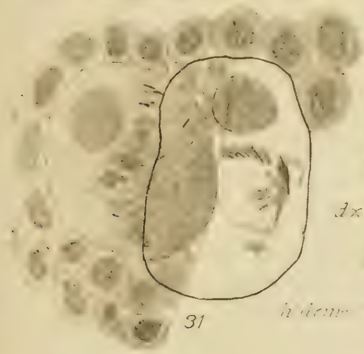

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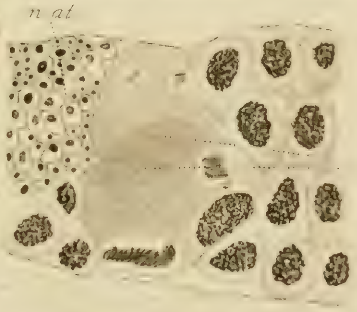

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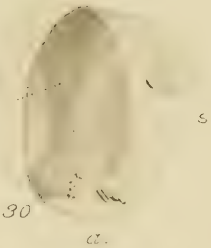

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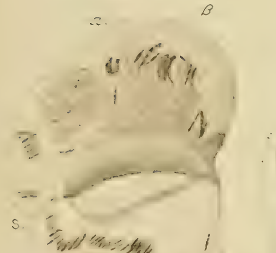

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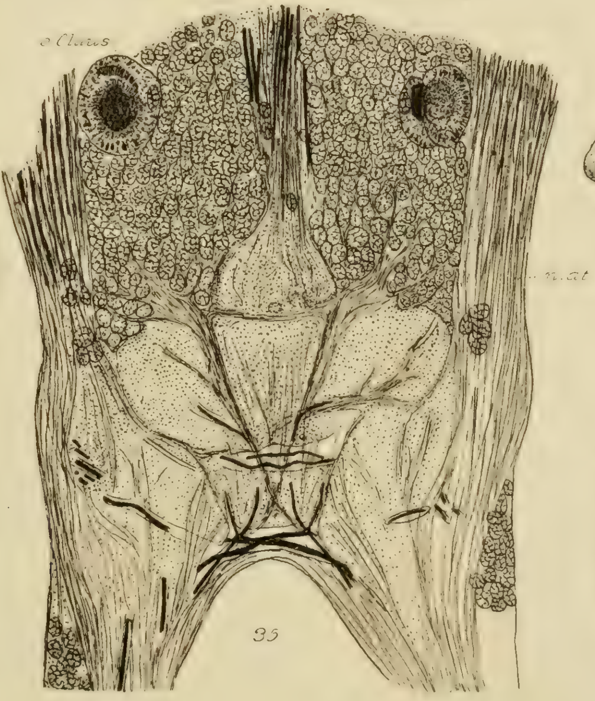

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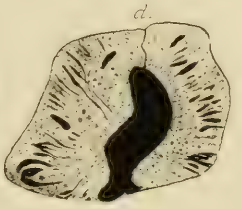

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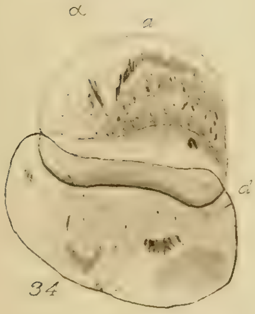

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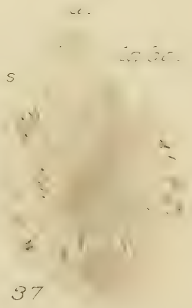

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C.O.E. del.

PLATE 4.

All the figures on this plate are cross sections from vom Rath's material and are magnified 1330 diameters; they are to show the number (28) of fibres in the optic nerve. Both frontal nerves (with sharp dark outlines) are also to be seen in each section. Compare Figure 27 (Plate **3**).

Fig. 38. Compare with Figure 1 (Plate **1**). The frontal nerves are still separated from the bundle of optic nerve fibres.

Fig. 39. A section a few sections posterior to that of Figure 38.

Fig. 40. Represents a section about midway between the eye and brain.

Fig. 41. Only a few sections separate this one from the anterior end of the brain.

Figs. 42, 43. Cross sections through the anterior part of the brain, from another series than that of Figures 38–41.

Figure 42. From a section just within the brain.

Figure 43. Somewhat posterior to Figure 42.

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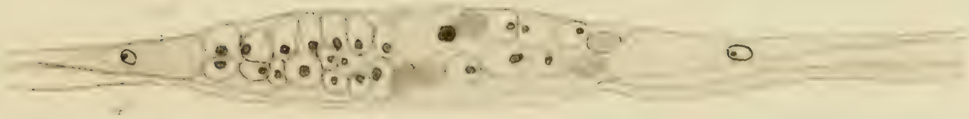

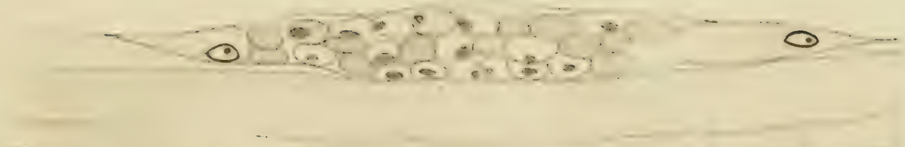

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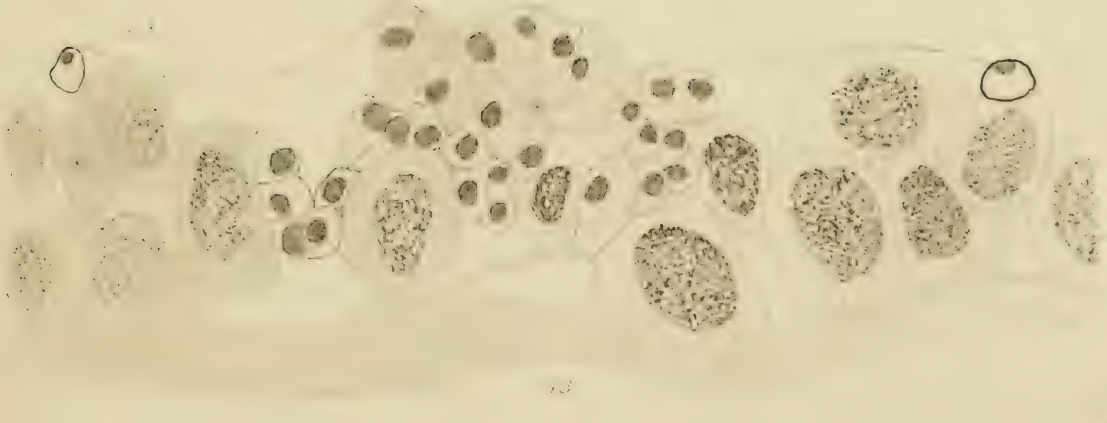

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C. O. E. del.

PLATE 5.

All the figures on this plate were drawn from preparations stained in Mallory's connective-tissue stain. It has been impossible to preserve satisfactorily in the monochromatic reproduction the desired values of the colored drawing.

Fig. 44. Oblique sagittal section, seen from right side. Note the membrane about the optic nerve; it is continuous with the membrane about the brain (compare with Fig. 43). The eye rests upon the ventral body wall; the cells of the ventral ocellus are continuous with the hypodermis. The membranes which are believed to belong only to the lateral ocelli are shown at the anterior end of the dorsal portion of the eye. The nerves lie in the median plane in the pigment cell; some fibres may be seen passing through the basal plate of the median ocellus. Neurofibrillae are also shown. × 830.

Fig. 45. Anterior face of a cross section through the posterior border of the eye. Twenty-eight fibres of the optic nerve are shown, two of which run through the basal plate of the ventral ocellus.

Fig. 46. Similar to Figure 44. In addition, two fibres of the optic nerve are shown passing from the retinal cells into the central cell at the anterior end of the dorsal portion of the eye. × 830.

Fig. 47. Cross section of the optic nerve. Note the sheath, and a nucleus; also the hypodermal nuclei. × 830.

Fig. 48. Sagittal section seen from left side. Compare with Figures 44 and 46. × 830.

Fig. 49. A cross-section, immediately anterior to that of Figure 45. Thirteen optic fibres are cut in cross sections, and six leave the cells of the lateral and ventral ocelli. The neurofibrillae, especially in the lateral portions of the eye, may be seen to be directly continuous with the nerve fibre. The club-shaped distal enlargements of the neurofibrillae are in immediate relation to the interior bodies — this being shown best in the ventral ocellus. × 830.

Fig. 50. The next cross-section anterior to that of Figure 49. The structures in the two are practically the same. But note in Figure 50 the opening in the basal plate of the ventral eye through which a nerve fibre passes. It is shown cut lengthwise in figures 44 and 48. × 830.

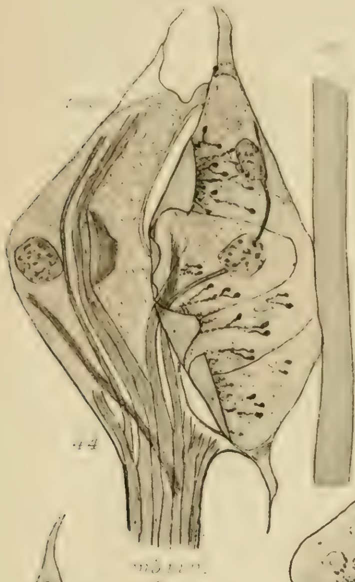
Fig. 51. Sagittal section of part of the brain and an organ of Claus. The membrane around the brain is shown, and the hypodermis beneath it. A nerve leaves the organ. All the structures — interior bodies, nucleus, nerve — are similar to those in the median eye. × 830.

FIG. 52. Three cells of the digestive tube, to show the striations in the basal portion. Compare with Figure 9 (Plate 1). × 830.

FIG. 53. From an oblique frontal section of the eye seen from dorsal side, showing that nerves leave the inner ends of the cells to make up the optic nerve. × 830.

FIG. 54. A cross section through the anterior part of the brain, to show the 28 fibres of the optic nerve within the brain. There is a membrane on the dorsal and one on the ventral surface of the brain; the latter separates the brain from the hypodermis. × 830.

FIG. 55. The next section to Figure 51. The basal plate and a row of interior bodies are shown. Two fibres leave the cell from a point on the periphery opposite to the basal plate. × 830.



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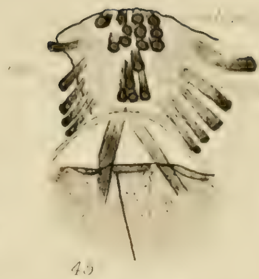

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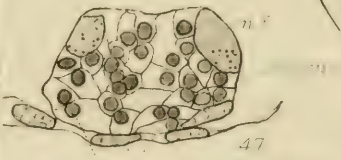

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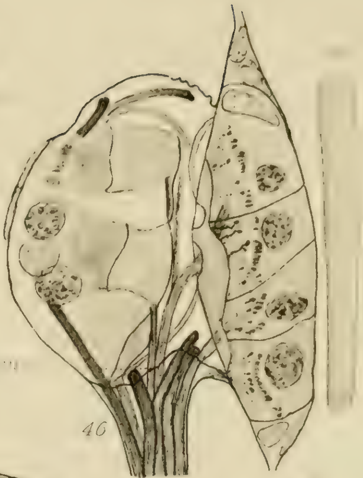

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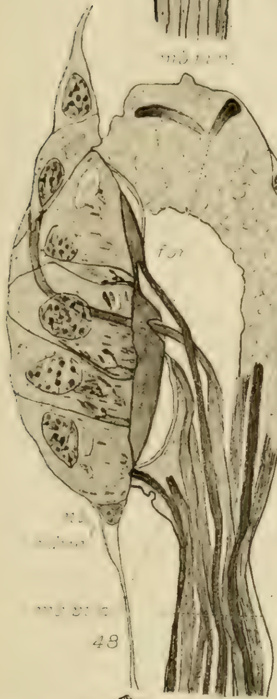

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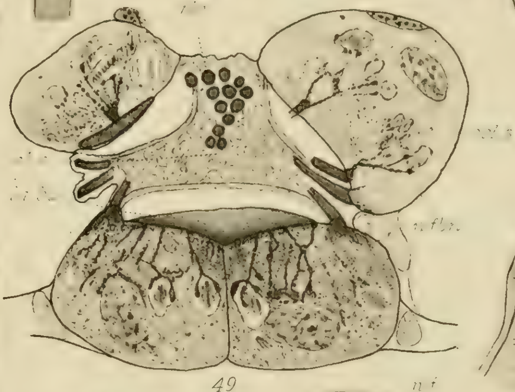

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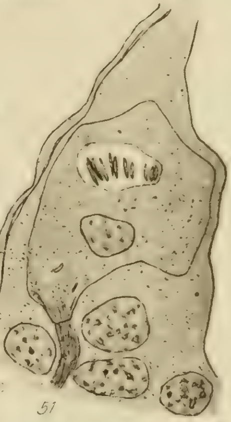

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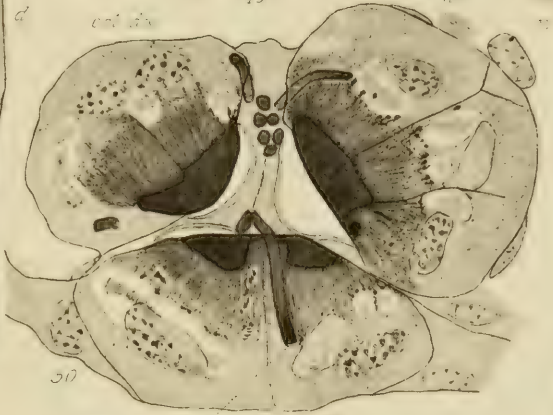

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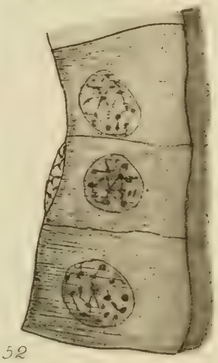

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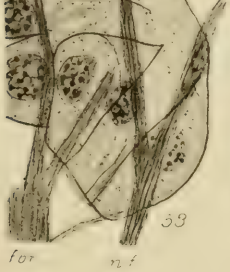

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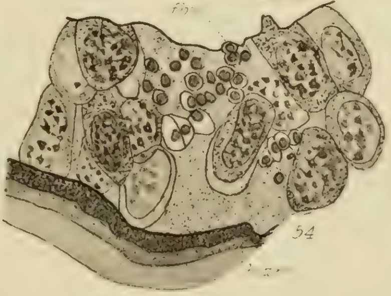

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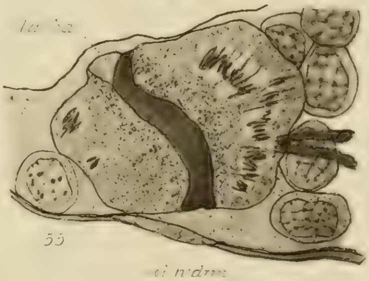

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C.O.E. del.

PLATE 6.

All figures reproduced from photographs of sections.

Fig. 56. Dorsal aspect of portion of frontal section of brain, showing the nerves (*n.*) from the organs of Claus. × 540.
The same section at slightly different focus and less magnification is shown in Figure 60.

Fig. 57. An organ of Claus in frontal section; basal plates and interior bodies (*pha'so.*) well shown. Vom Rath. × 405.

Fig. 58. The section shown in Plate 1, Figure 7. × 380.

Fig. 59. The section drawn in Figure 2 (Plate **1**).

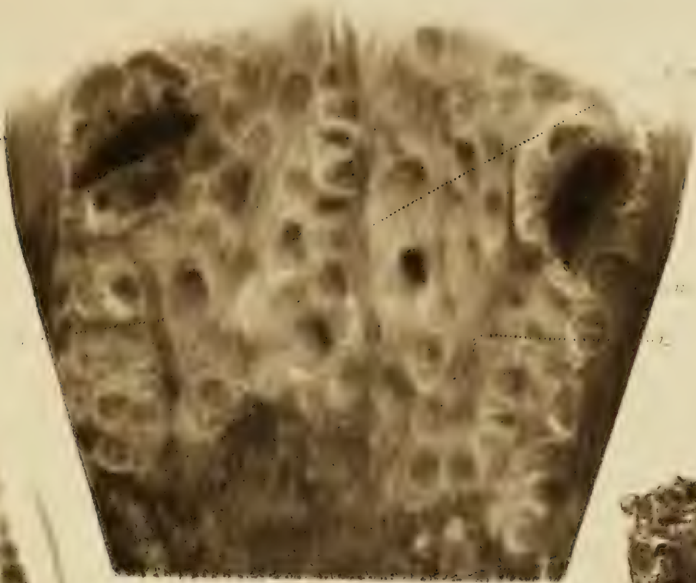
Fig. 60. Dorsal aspect of portion of frontal section of brain, showing the organs of Claus. × 390.

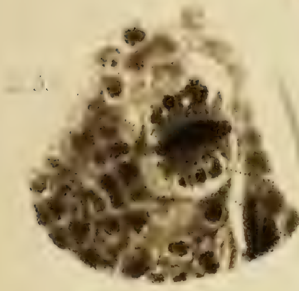
Fig. 61. The section from which Figure 18 (Plate **2**) was drawn. × 420.

Fig. 62. Frontal section of eye seen from the dorsal side. Vom Rath. × 475.

Fig. 63. Anterior face of a cross section of an eye, showing sharply defined basal plates, tapetum, hypodermis, and cuticular body wall. Zenker; iron haematoxylin. × 560.

Fig. 64. The section immediately dorsal to the one shown in Figure 59. × 500.





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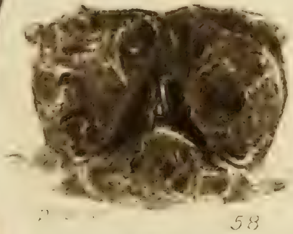

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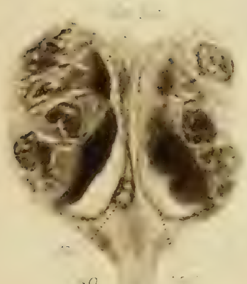

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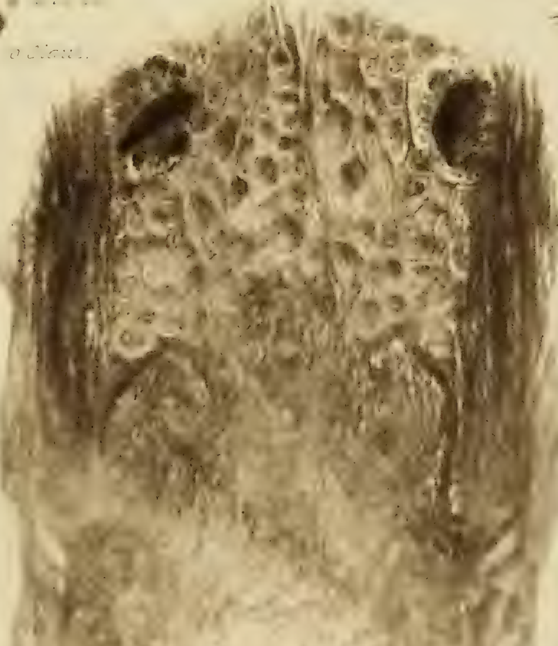

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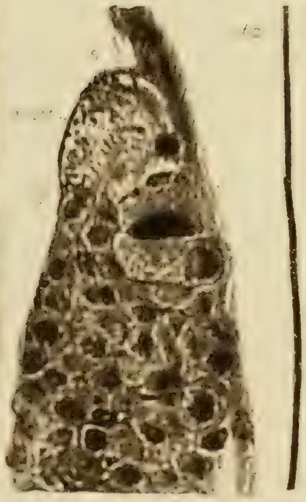

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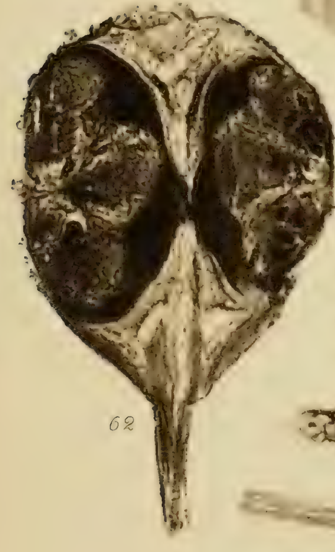

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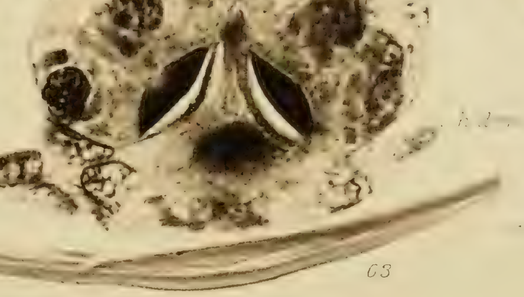

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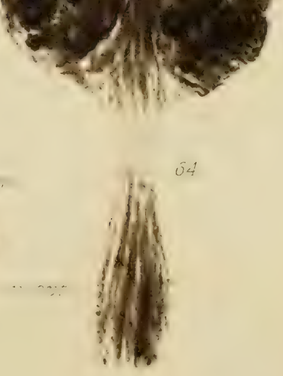

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Bulletin of the Museum of Comparative Zoölogy
AT HARVARD COLLEGE.
Vol. LIII. No. 2.

SPERMATOGENESIS IN ACRIDIDAE AND LOCUSTIDAE.

By Herbert Spencer Davis.

With Nine Plates.

CAMBRIDGE, MASS., U. S. A.:
PRINTED FOR THE MUSEUM.
December, 1908.

No. 2.—CONTRIBUTIONS FROM THE ZOÖLOGICAL LABORATORY OF THE MUSEUM OF COMPARATIVE ZOÖLOGY AT HARVARD COLLEGE, UNDER THE DIRECTION OF E. L. MARK, No. 197.

Spermatogenesis in Acrididae and Locustidae.

By Herbert Spencer Davis.

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I. Introduction.

The investigations which form the basis of this paper were begun in the Zoölogical Laboratory of Harvard University during the winter of 1900–1901. They were continued at irregular intervals in connection with the author's work at the Washington State College, Pullman, Wash., and have been brought to completion during the present year (1906–1907) in the Zoölogical Laboratory of Harvard University. However, most of the observations can be considered as made during the present year since, in addition to much new material studied, the previous work has been gone over anew and most of the figures redrawn. My warmest thanks are due to Prof. E. L. Mark, under whose direction this work has been done and to whom I am indebted for many helpful suggestions and criticisms.

Considerable work has been done upon the spermatogenesis of the Orthoptera but with remarkably diverse results. There is probably no other group in which such radically opposed conclusions have been reached by different investigators. Just why this is so, it is hard to say. The material is in general very favorable for cytological work, since the cells and chromosomes are large, and in many species exhibit remarkably clear structures.

Carnoy ('85) was the first to describe and figure the divisions of the male germ cells of Orthoptera. Von La Valette St. George ('86) described the spermatocyte divisions and the metamorphosis of the spermatid in Blatta. Vom Rath ('92, '95) found that in the spermatogenesis of the mole cricket (Gryllotalpa) the first maturation division was longitudinal, the second transverse, or in other words a reduction division. On the other hand, Wilcox ('95, '96), working on Caloptenus (Melanoplus, an acridid), arrived at very different conclusions, holding that in this species both maturation divisions are transverse. Wilcox also gave a detailed account of the metamorphosis of the spermatid. In 1899 McClung in a short paper called attention to the existence in the male germ cells of Xiphidium of a peculiar chromosome, characterized by remaining compact and staining deeply during the resting stage of the primary spermatocyte. This element he called the "accessory chromosome." In a later paper McClung (:00) gave a detailed account of the spermatocytes and the maturation divisions in Hippiscus (one of the Acrididae). He concluded that the first maturation division is longitudinal, while the second is transverse, and that the accessory chromosome, like the other chromosomes, divides at both divisions. In the same year Sutton (:00) found that in the spermatogonia of Brachystola (one of the Acrididae) the chromosomes exhibit a remarkable degree of separation, for during the telophase of the secondary spermatogonial divisions each chromosome becomes enclosed in a distinct vesicle. Later these vesicles fuse at their polar extremities, with the exception of the one containing the accessory chromosome, which remains distinct throughout the resting stage.

In 1901 de Sinéty, in an extended paper on the anatomy and physiology of the Phasmidae, published an account of the spermatogenesis of a number of Orthoptera belonging to several families. He concluded that both maturation divisions are longitudinal or equational, but that the accessory chromosome does not divide in the first division. Thus only one half of the spermatids contain this chromosome. A little later McClung (:02[a]), in an account of the spermatocyte divisions of the Locustidae, confirmed de Sinéty's account of the behavior of the accessory chromosome during these divisions. McClung also showed that during the spireme stage of the spermatocytes the accessory chromosome becomes converted into a long coiled thread. His account of the maturation divisions agrees with that previously given by him for the Acrididae. Sutton (:02) in an interesting paper showed that in the spermatogonia of Brachystola the chromosomes vary greatly in size, but that with one exception, the accessory, there are always two of each size, and that in the spermatocytes the bivalent chromosomes have the same size relations. He believed that the bivalent chromosomes were formed by the conjugation end to end of the individuals of each pair of the spermatogonia. Sutton agreed with McClung, that the second maturation division is the reducing division.

In 1902 Baumgartner published an account of the metamorphosis of the spermatid in Gryllus, and later (:04) showed that in the spermatogonia the accessory chromosome forms a large V-shaped structure, very different from the ordinary rod-shaped chromosomes. In the growth stages of the spermatocytes the accessory chromosome forms, as in the other Orthoptera, a deeply staining mass applied to the nuclear membrane, and divides only during the second maturation division. Baumgartner also believed that in the spermatocytes there is evidence that the ordinary chromosomes have individual morphological characteristics.

In striking contrast to the results of McClung and Sutton, Montgomery (:05) concluded that in Syrbula (an acridid) the first maturation division instead of the second, is the reducing division. He also found that the accessory chromosome of the spermatocyte is represented in the spermatogonia by two chromosomes, and that it divides during the maturation divisions in the same way as the other chromosomes. Farmer and Moore (:05) also held that in the cockroach (Periplaneta) the first maturation division is the reducing division. Moore and Robinson (:05) have reached the conclusion that the so called accessory chromosome in Periplaneta is nothing more than a true nucleolus, and that it disappears before the first maturation division. On the other hand Stevens (:05^a) has found that in Blatella the accessory chromosome has practically the same history as that described by McClung, Sutton, and de Sinéty for other Orthoptera. However, in Stenopelmatus (the California sand cricket) she found a structure in the primary spermatocyte which, in position and form, resembled the accessory chromosome, but differed from it in mode of origin. During the first maturation division this element passes bodily into one of the daughter cells, where it degenerates before the second division.

Some very surprising results have been reached by McClung (:05), who believes that in several species of the Acrididae and in one locustid (Anabrus) there is, even in the spermatogonia, a bivalent chromosome to the end of which the accessory chromosome becomes attached during the prophase of the first spermatocyte division. In one species the chromatic element formed by the union of a bivalent and the accessory chromosomes unites with another bivalent chromosome so that there is a single element composed of two tetrads and the accessory chromosome. During the first maturation division one entire tetrad and the accessory pass into one, the other tetrad into the other daughter cell.

Recently Otte (:06) has arrived at results diametrically opposed to those of most of the preceding investigators. He finds that in the spermatogonia of Locusta the chromosomes remain perfectly distinct during the resting stage, each chromosome lying in a separate vesicle. During the early growth stages of the primary spermatocytes the chromosomes are in the form of chromatic filaments, which conjugate, side by side, in pairs. The bivalent chromosomes thus formed are divided transversely in both maturation divisions, there being no reduction in Weismann's sense. Otte agrees with McClung, that during the growth period of the primary spermatocyte the accessory chromosome forms a spireme, but thinks that it divides transversely during the second division. Otte (:06[a]) also gives a detailed account of the metamorphosis of the spermatid.

In a recent paper Gutherz (:07) confirms, in the main, Baumgartner's observations on the accessory chromosome in Gryllus.

II. Material and Methods.

The following investigations are based on material from seven species of Orthoptera belonging to two families, although a number of other species have been examined from time to time for comparison. Six species of the Acrididae have been selected for description, as follows: Dissosteira carolina, Arphia tenebrosa, Hippiscus tuberculatus and Chortophaga viridifasciata, belonging to the subfamily Oedipodinae; Melanoplus femoratus, belonging to the subfamily Acridinae; and Stenobothrus curtipennis, belonging to the subfamily Tryxalinae. In addition an account is given of the spermatogenesis of Steiroxys trilineata, a locustid.

The material was obtained mostly from adult individuals collected in three localities, as follows:—Chortophaga viridifasciata and Hippiscus tuberculatus at Cambridge, Mass.; Dissosteira carolina, Melanoplus femoratus, and Steiroxys trilineata at Pullman, Wash., while Stenobothrus curtipennis was collected at Cambridge, Mass., Pullman, Wash., and Torrington, Conn.

It has seemed best to describe the spermatogenesis of one species as completely as possible, and in the other forms, in order to avoid needless and tiresome repetition, to consider only points which are of especial interest. The most complete account is given for Dissosteira carolina, not because the material was more favorable than in some of the other species, but because it is a common form and is considered representative of the subfamily to which it belongs.

In all cases the testes were dissected out in a 0.6% salt solution and placed at once in the fixing fluid. A number of fixing fluids were tried, namely: Hermann's platino-aceto-osmic, Flemming's chromo-aceto-osmic (strong formula), Worcester's formol-sublimate-acetic mixture, and Zenker's bichromo-sublimate-acetic mixture. It was found that much the best fixation was obtained with Hermann's fluid; consequently this was later used almost exclusively, although Flemming's fluid was used in a few cases. In using both these fluids, the testes were left in the fluid from one to two hours. All the material, after being hardened a few days in alcohol was imbedded in paraffin and kept in that condition until sectioned. Sections were cut from three to ten micra thick. A number of staining methods were tried, but it was soon found that Heidenhain's iron hematoxylin preceded by Bordeaux R, gave results which were much superior to any other method, and in the later part of the work this stain was used almost exclusively. In using this method the best results were obtained by staining in a 1% aqueous solution of Bordeaux for twenty-four hours, then transferring to the ferric alum for from one to two hours, after which the sections were placed in hematoxylin for from four to six hours. It is necessary to dehydrate very rapidly, as the Bordeaux is quickly extracted by the alcohol, but with a little care the desired intensity of the cytoplasmic stain can be obtained. Hermann's safranin-gentian combination was used in some cases, but was found to be unreliable as a differential stain for the monosome, as I shall call the accessory chromosome.

Most of the terms employed are those in common use among cytologists, and so require no explanation, but for the sake of clearness and simplicity it has seemed best to adopt in the case of the chromosomes the nomenclature proposed by Montgomery (:06) as follows: *Chromosome,* the word introduced by Waldeyer, to be used as a general term for each separate mass of chromatin and linin which appears in the cell during mitosis. *Autosomes,* the non-aberrant chromosomes. *Allosome,* any chromosome which behaves differently from the autosomes. Two kinds of allosomes are distinguished as follows:

(1) *Monosome.* Allosomes which are unpaired in the spermatogonium. These chromosomes have been called variously, accessory chromosomes (McClung), chromosomes speciaux (de Sinéty), heterotropic chromosomes (Wilson), etc.

(2) *Diplosome.* Allosomes which are paired in the spermatogonia. These have been called chromatin nucleoli (Montgomery), idiochromosomes and m-chromosomes (Wilson), and heterochromosomes (Stevens).

III. Observations.

1. Structure of the Testis.

The following description of the testis, although based primarily on Dissosteira carolina, will apply equally for all the Acrididae, since the structure is essentially the same throughout the family.

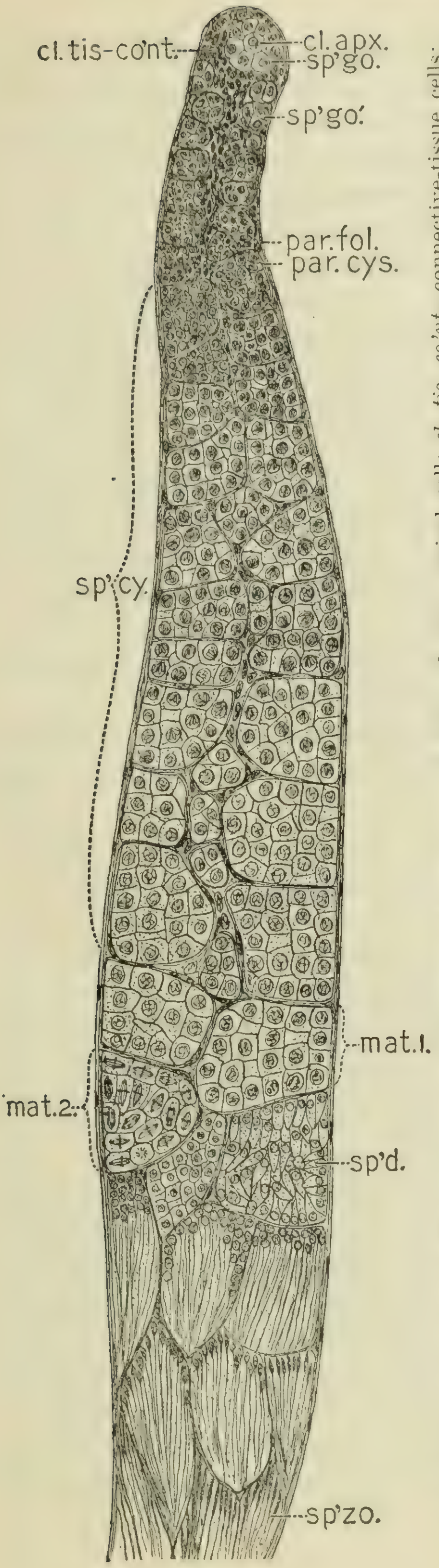


FIG. *A.*— Longitudinal section of follicle of Dissosteira carolina. × 100. *cl. apx.*, apical cell; *cl. tis. co'nt.*, connective-tissue cells; *mat. 1.*, prophase first maturation division; *mat. 2.*, second maturation division; *par. cys.*, cyst wall; *par. fol.*, follicular wall; *sp'cy.*, primary spermatocytes, growth period; *sp'd.*, spermatids; *sp'go.*, primary spermatogonia; *sp'go'.*, secondary spermatogonia; *sp'zo.*, spermatozoa.

The testes, which are so closely apposed as to present the appearance of a single organ on superficial examination, lie in the dorsal part of the fourth to the sixth abdominal segments. Each testis is composed of a number of elongate, cylindrical follicles (apparently corresponding to ovarioles), tapering somewhat at either end and lying approximately parallel to each other. However, in adults after the spermatozoa have partially matured the proximal ends of the follicles become much constricted. Each follicle is connected at its anterior (proximal) end with the vas deferens. The follicles of each testis are inclosed in a delicate connective-tissue membrane, which usually contains an orange colored pigment.

In a longitudinal section each follicle (Fig. *A*) is seen to be filled with a mass of germ cells in successively older stages from the distal to the proximal end. At the distal end there is always a single large apical cell around which the primary spermatogonia are arranged in a single layer. Surrounding the primary spermatogonia are large numbers of connective-tissue cells, which are easily distinguished by their relatively small size and deeply staining nuclei. These cells extend proximally along the center of the follicle for some distance, forming a more or less distinct longitudinal rachis. Proximal to the primary spermatogonia are the secondary spermatogonia arranged in groups, which are surrounded by a thin but distinct membrane, composed of connective-tissue cells, each group constituting a spermatocyst. Each spermatocyst remains distinct from its fellows from the time of its formation until the spermatozoa become mature. The spermatocysts are arranged in regular sequence from the distal to the proximal end of the follicle, so that successively older stages in the development of the germ cells are encountered as one goes toward the proximal end. This is, of course, due to the fact that existing cysts are continually being forced toward the vas deferens by the formation and growth of new cysts at the distal end; thus cysts which are formed at about the same time come to occupy neighboring positions in the follicles.

Surrounding the whole is a distinct follicular wall, in which flattened, deeply staining nuclei can be distinguished at intervals.

In Steiroxys the structure of the testis is much the same, except that in the later stages a single spermatocyst may come to occupy the entire cross-section of a follicle.

2. The Spermatogonia.

A. Dissosteira carolina.

The primary spermatogonia (Plate 1, Fig. 5) are large rounded cells situated at the distal end of the follicle. Each cell contains a large eccentrically placed nucleus, which is usually more or less irregular or lobulate in shape, the nuclear membrane often extending out in the form of short sacculations, like the fingers of a glove. This irregular or polymorphic form of the nucleus is more marked in the secondary spermatogonia, where it will be considered more at length. On one side of the primary spermatogonia the cytoplasm is much more abundant than elsewhere, and adjacent to this cytoplasmic mass the nucleus is flattened or concave. At the height of the resting stage the chromatin is distributed through the nucleus in the form of fine granules suspended in the linin network. At intervals, especially at the intersections of the linin threads, the chromatin granules tend to aggregate in irregular masses. In addition, the nucleus contains one, or in some cases two, small, rounded plasmosomes, which are usually stained with the Bordeaux red. However, if the preparations are not strongly decolorized the plasmosomes may retain the iron hæmatoxylin.

As stated above, the cytoplasm is especially abundant on one side of the cell, and in this region there can be distinguished a rather diffuse very finely granular material, staining more deeply with Bordeaux than the surrounding cytoplasm. This apparently corresponds to the mitochondrion of Benda.

The primary spermatogonia divide exclusively by mitotic division, the process being the same as in the secondary spermatogonia, where it will be described in detail.

The arrangement of the primary spermatogonia is characteristic. They are always grouped in a single layer about a peculiar cell located at the distal end of the follicle. This cell appears to be homologous with the *apical*, or *Verson's cell* described by a number of investigators in the testes of various insects. There is always one of these cells in each follicle, and it is surrounded on all sides by a layer of primary spermatogonia so arranged that the sides containing the most cytoplasm and the mitochondrion are in contact with it. The apical cell (Fig. 1) is usually larger than the primary spermatogonia, irregularly polygonal and sometimes sends out short processes between the surrounding spermatogonia. Within the cell is a large, eccentrically located nucleus, through which the chromatin is distributed in flocculent masses. These masses are composed largely of a chromatic material or linin in which fine chromatic granules are embedded. The linin also extends through the nucleus in somewhat coarse strands connecting the chromatic masses with each other and with the nuclear membrane. Suspended in the nuclear network are several irregularly shaped plasmosomes, which apparently are simply masses of linin.

Directly surrounding the nucleus the cytoplasm contains a large quantity of a more or less distinctly granular material staining with the plasma stain. This material is especially abundant on one side of the nucleus, but extends as a thin layer almost entirely around it. In this finely granular mass there are usually large numbers of larger, rounded granules staining deeply with iron hematoxylin. In some cases these granules are very abundant, while in others they may be almost entirely absent. A careful study of a large number of these cells has shown that there are all transitions between the two extremes, and that the finely granular material is probably derived from larger deeply staining granules, which gradually lose their affinity for hematoxylin and break down into very fine granules.

The cytoplasm outside the granular mass is abundant, but much clearer than that of the surrounding spermatogonia.

The study of a large number of testes of different ages in this and other species has failed to show any other stages in these cells. They always appear approximately the same except for the differences in the condition of the granular mass surrounding the nucleus.

Lying outside and surrounding the primary spermatogonia are large numbers of connective-tissue cells (Plate 4, Fig. 67), which can be easily distinguished from the spermatogonia by their smaller size, the relatively small amount of cytoplasm, and the deeper staining qualities of the nucleus. When the primary spermatogonia divide, one of the daughter cells is usually forced out of the layer of cells surrounding the apical cell. This cell then becomes intimately associated with one or more of the connective-tissue cells, which form an investment around it. This investment persists until the descendants of the cell are converted into spermatozoa.

Sutton (:00, :02) maintained that in Brachystola the spermatogonia and the cyst cells have a common origin, and that in the earlier generations they are indistinguishable. I believe that the cells which he at first (:00) took to be primary spermatogonia and later (:02) to be the first generation of secondary spermatogonia are all, in reality, connective-tissue cells. Sutton was unable to find any transitional stages connecting these cells with the later generations of spermatogonia. In my own preparations of Dissosteira and other species of the Acrididae, the two types of cells are always easily distinguishable, and I have seen nothing to indicate a genetic connection.

The investment of spermatogonia by cyst cells marks the transition from primary to secondary spermatogonia, the spermatogonium with the surrounding cyst cells forming the beginning of a spermatocyst. The secondary spermatogonia divide mitotically in rapidly succeeding divisions, the daughter cells remaining enclosed by the cyst wall. It is thus evident that all the cells in each cyst are the direct descendants of a single primary spermatogonium, which became surrounded by a connective-tissue investment. At the distal end of each follicle are always a number of cysts containing from one or two up to a large number of secondary spermatogonia, according to their age. In striking contrast to the primary spermatogonia, where adjacent cells are often in very different stages, all the cells of a cyst are usually in practically the same stage of development.

Occasionally nuclei in the cyst wall can also be seen undergoing mitosis.

The appearance of the secondary spermatogonia in all generations is practically the same except in size, the later generations being much smaller than the earlier. This is, of course, due to the fact that the growth of the cells does not keep pace with the divisions, which succeed each other rapidly. There is, however, a considerable growth, since cysts containing the later generations of spermatogonia are much larger than those composed of earlier generations. The secondary spermatogonia of the earlier generations are even larger than the primary spermatogonia and the amount of cytoplasm is usually relatively greater (Plate **4**, Fig. 68). In a section through one of the smaller cysts the spermatogonia are seen to have a radiate arrangement around a common center. Each cell is roughly conical with the nucleus eccentrically situated in the broad base near the cyst wall. Extending through the cytoplasm at the apex of each cell, and connecting it with one or more adjoining cells, can usually be distinguished a very finely granular, nearly homogeneous mass staining deeply with Bordeaux. This is the remnant of the interzonal filaments, which often persist for several generations, although naturally those of the last division are most prominent. Following Mark and Copeland (:**06**), I shall speak of these as the *interzonal bodies*. In addition to the interzonal bodies there is in the cytoplasm of this part of the cell a more or less diffuse finely granular material staining with Bordeaux, which is apparently to be considered mitochondrion. It is never very abundant or conspicuous in the spermatogonia of this species, and in some cases it is almost impossible to distinguish it at all.

In the later generations the spermatogonia lose this regular arrangement and the cysts are seen to be made up of large numbers of irregularly arranged, rounded or polygonal cells crowded closely together; all being connected with one another by means of the persistent interzonal bodies.

Usually the cells of each cyst are all in approximately the same stage, but in the later generations rarely the cells of one side of a cyst may be considerably in advance of the others.

Turning now to a more detailed description of the secondary spermatogonia, we find that during the so-called resting stage the nuclei have practically the same structure as in the primary spermatogonia, except that they are more chromatic, especially in the later generations, and often more irregular in shape (Plate **1**, Fig. 6). There is no continuous spireme formed in preparation for division, but the chromatin becomes aggregated into a number of long, much convoluted threads, which rapidly shorten and thicken into contorted rods of varying lengths (Figs. 7, 8). These rods at first have a ragged outline and often show a more or less distinct longitudinal split. Frequently the ends of the rods can be traced out into the sacculations of the nuclear wall. In sections through one side of the nucleus (Figs. 9, 10) the rods often appear to lie in distinct compartments separated from each other by a thin membrane. This, however, is probably not the case. Apparently these compartments are simply outpocketings or sacculations of a common nuclear membrane which have been cut in such a way as to give the appearance of distinct vesicles. The chromatic rods continue to shorten and thicken and become more compact, each one forming a single rod-shaped chromosome. Shortly after the chromatic rods appear in the nucleus, there can be distinguished in the cytoplasm close to the nuclear membrane two small but well defined asters (Figs. 7, 8). At the center of each aster there is usually a very minute deeply staining granule, which is evidently the centrosome. It is, however, doubtful if these centrosomes are present when the asters first appear, for, in a number of cases where the asters were barely distinguishable, I have been unable to find any centrosomes at the center. The centrosomes when they first appear are very minute, but increase rapidly in size. The asters always make their appearance on that side of the nucleus which is directed towards the interzonal body, and is much flattened or even concave. Following Montgomery (:00, p. 294) I shall call this the distal pole, the opposite one being the central pole of the cell.

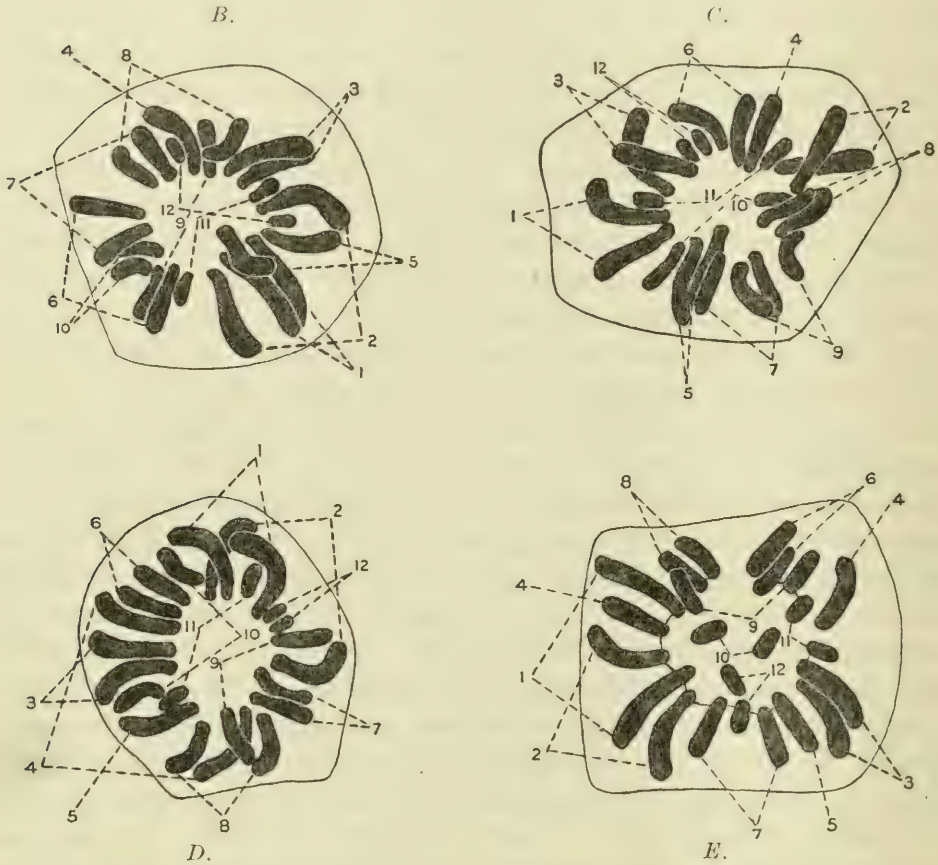
Whether the centrosomes retain their individuality during the resting stage, it is impossible to say, although the manner of their appearance during the prophase would argue against such an interpretation. If they do exist as morphological entities in the resting cell, they must be so minute as to be indistinguishable. Since in the prophase the centrosomes reappear at the distal pole while during the telophase of the preceding division they were at the central pole, it is obvious that their persistence during the resting stage involves their migration from the central to the distal pole through, or around the nucleus.

When the two asters first appear they are always some distance apart, and in no case have I been able to distinguish a central spindle or centrodesmus between them at this stage. However, they may be connected to each other by the astral rays which extend out for a considerable distance in all directions. Surrounding the asters is a mass of mitochondrion from which the astral rays appear to be formed, but it is impossible to determine this with any certainty. As the chromosomes shorten and thicken, the asters and centrosomes increase rapidly in size and become connected by a well defined central spindle. At the same time the nuclear membrane gradually disappears, first disintegrating on the side adjacent to the asters. Figure 11 shows a somewhat later stage, in which the nuclear membrane has entirely disappeared and the spindle is moving in among the chromosomes, which are connected at one end with the astral rays. At a little later stage (Fig. 13) the spindle is much larger and is composed of two types of fibers, the coarser mantle fibers attached to the chromosomes and the finer central fibers. The chromosomes are now arranged in a single plane around the periphery of the spindle and are attached at one end to the mantle fibers. At this stage the centrosomes reach their greatest development and can be distinctly seen as large deeply staining bodies at the poles of the spindle. The astral rays are now few and indistinct. At no time during mitosis is there any sign of a centrosphere around the centrosome.

The chromosomes now all divide longitudinally and pass to the poles of the spindle, as in ordinary mitotic division (Fig. 14).

Cross sections of the spindle in the metaphase (Figs. *B*, *C*, and Plate **1**, Fig. 12) show in favorable cases twenty-three rod-shaped chromosomes arranged in a ring around the periphery of the spindle. These chromosomes vary greatly in size and a careful study shows that they form a nicely graded series from the largest to the smallest. It is also evident that, as has been shown by a number of recent investigators in other insects, the gradations in volume are not between single chromosomes, but between pairs of chromosomes. In Figures *B* and *C* I have attempted to indicate the members of each pair, although on account of fore-shortening and the slight difference in volume in some cases no pretence is made to complete accuracy. There are three large pairs 1, 2 and 3 (Figs. *B*, *C*). The next chromosome, 4, is without a corresponding mate and is therefore the monosome. Following this the pairs 5, 6, 7, 8, 9, 10 form a series constantly diminishing in size. There is a marked break in size between 10 and the larger of the two smallest pairs, 11 and 12. (The different pairs are much more clearly marked in Stenobothrus, where there is a greater difference in size.) There are thus in the spermatogonia twenty-two autosomes and one monosome.

Figure 14 (Plate **1**) shows the anaphase, Figure 15 the telophase of the spermatogonial division. In the telophase as the chromosomes begin to break down the projecting end of each is usually enclosed in a distinct vesicle. It is, however, probable that these vesicles are continuous at the central pole and that at no time is there a distinct vesicle for each chromosome. It is impossible to distinguish the monosome with certainty during this stage as Sutton (:00, :02) has been able to do in Brachystola, but in some cases one of the chromosomes has a somewhat different appearance and is surrounded by a more distinct vesicle. Judging from Sutton's work, this is probably the monosome, but in this species the difference from the other chromosomes is not great enough to render it distinguishable in most cases. However, even at the height of the resting stage there can sometimes be recognized a more or less distinct vesicle in which the chromatin is more densely aggregated than elsewhere. This vesicle with its more deeply staining chromatin usually extends out into the cytoplasm near



Figs. *B.–E.*— Polar views, metaphase of spermatogonia showing autosome pairs. × 1450.
Figs. *B* and *C.*— *Dissosteira carolina.*
Fig. *D.*— *Arphia tenebrosa.*
Fig *E.*— *Chortophaga viridifasciata.*

the middle of the concave side of the nucleus. This is of considerable interest, since in the early stage of the primary spermatogonia the monosome usually occupies a similar position.

As the chromosomes disintegrate during the telophase they are, in general, oriented with their long axes parallel to a line extending through the two poles of the cell. During the resting stage all traces of chromosomal limits disappear, but in the succeeding prophase, when the chromosomes reappear, they have the same orientation as in the telophase of the preceding division. Obviously, the most plausible explanation of this is that during the resting stage the chromosomes still retain their individuality, although they are not recognizable as distinct bodies. Otherwise, it is difficult to see why they should reappear with the same orientation as before. Somewhat similar phenomena have been described by Rabl ('85) and Boveri ('88).

B. Arphia tenebrosa.

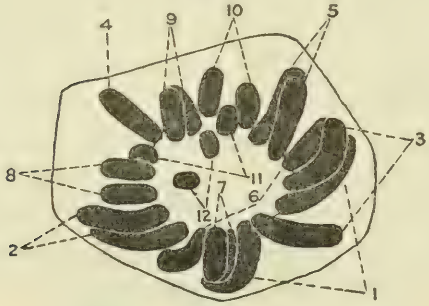
The spermatogonia and apical cell are approximately as in Dissosteira. Figure *D* (p. 72) is a polar view in the equatorial-plate stage showing, as in Dissosteira, twenty-three chromosomes. These can be easily arranged in pairs, but in this species there are four large pairs; the next smaller chromosome is unpaired, the monosome, and this is followed by a series of five nicely graded pairs of diminishing sizes. There is, as in Dissosteira, a greater difference in size between the smallest of the medium sized chromosomes and the two smallest pairs than between the successive sizes of the other chromosomes.

C. Hippiscus tuberculatus.

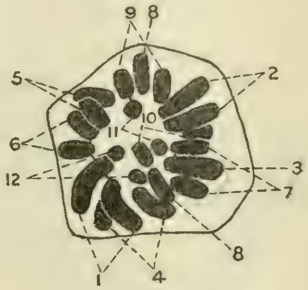
The spermatogonia present no essential differences from those of Dissosteira. The apical cell, however, differs somewhat in appearance, there being in most cases no deeply staining granules in the cytoplasm. Usually there is around the nucleus simply a very finely granular material, which is comparable to that seen in Dissosteira, but stains much less deeply. Moreover, the granular material is distributed equally on all sides of the nucleus. In this species the primary spermatogonia usually do not entirely surround the apical cell, which consequently is in contact on one side with connective-tissue cells.

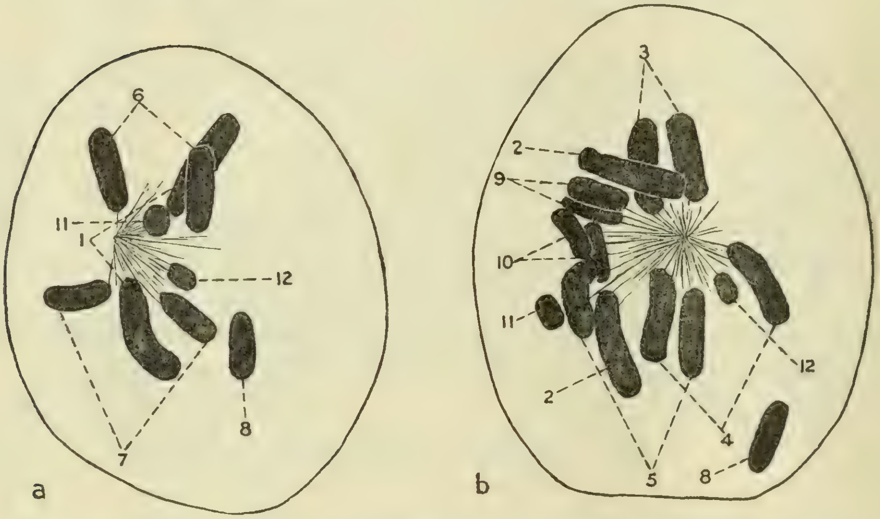
In the metaphase twenty-three chromosomes can be distinguished which, as in the preceding species, vary greatly in size. Figure *F*, which is a polar view in the equatorial-plate stage, shows that here, again, the chromosomes are evidently in pairs. Next in size to the three largest pairs (1, 2, 3) is an unpaired chromosome (4), the monosome. Then in a series of diminishing sizes we have the pairs 5, 6, 7, 8, 9, 10. As in the preceding species, there are two pairs, 11 and 12,

F.



G.





H.

FIG. *F.*— Polar view, metaphase of spermatogonium in *Hippiscus tuberculatus* showing autosome pairs. × 1450.

FIG. *G.*— Polar view, metaphase of spermatogonium of *Melanoplus femoratus* showing autosome pairs. × 1450.

FIGS. *Ha, Hb.*— Successive sections of oögonium of *Hippiscus tuberculatus* during prophase showing autosome pairs. × 1450.

which are much smaller than the rest. Figures *Ha* and *Hb* represent two successive sections of an *oögonium* of this species in the late prophase. Here there are plainly twenty-four chromosomes, all of which can be readily arranged in twelve pairs. A comparison of these with Figure *F* will show that these pairs correspond exactly with those of the spermatogonia, except that in place of the monosome (4) there is in the oögonia a symmetrical pair. This is in accord with the results of Wilson (:05 [b]), who has shown that in those Hemiptera in which a monosome occurs in the male there is always one more chromosome in the oögonia. Miss Stevens (:06 [a]) has found the same to be true of several Coleoptera. However, Sutton (:02) found only twenty-two chromosomes in the ovarian follicular cells of Brachystola, while in the spermatogonia there are twenty-three chromosomes.

D. Chortophaga viridifasciata.

The spermatogonia and apical cell are much as in Dissosteira. In a polar view of the equatorial-plate stage (Fig. *E*, p. 72) there are also twenty-three chromosomes, of which twenty-two are autosomes, since they can be readily paired. The remaining chromosome (4) is a monosome. I have been unable to find any evidence of a precocious conjugation of the spermatogonial chromosomes such as McClung (:05) found in this species.

During the prophase of the last spermatogonial division the monosome can be easily distinguished, since it has a much more ragged outline than the autosomes and lies in a more or less distinct vesicle. In the prophase of the other spermatogonial divisions it cannot be distinguished from the autosomes.

E. Melanoplus femoratus.

The testicular elements in Melanoplus are much smaller than in the preceding species, and, therefore, are not in general so favorable for investigation. The primary spermatogonia are much like those of Dissosteira and partially surround the apical cell. On its proximal side this cell is surrounded by connective-tissue cells. The apical cell (Plate 1, Fig. 3), like the other elements of the testis, is much smaller than in Dissosteira and contains no deeply staining granules in the cytoplasm, only the finely granular material which stains lightly with Bordeaux being present.

The secondary spermatogonia show no essential difference from the same cells in Dissosteira, but the nuclei are even more irregular in shape. During the resting stage, when the chromatin has become most widely diffused through the nucleus, there is often present a deeply staining mass formed by an aggregation of chromatin granules; whether it has any connection with the monosome, I am unable to say.

In the metaphase (Fig. *G*, p. 74) twenty-three chromosomes can be distinguished in the equatorial plate and, as in all the preceding species, the autosomes can be readily paired.

F. Stenobothrus curtipennis.

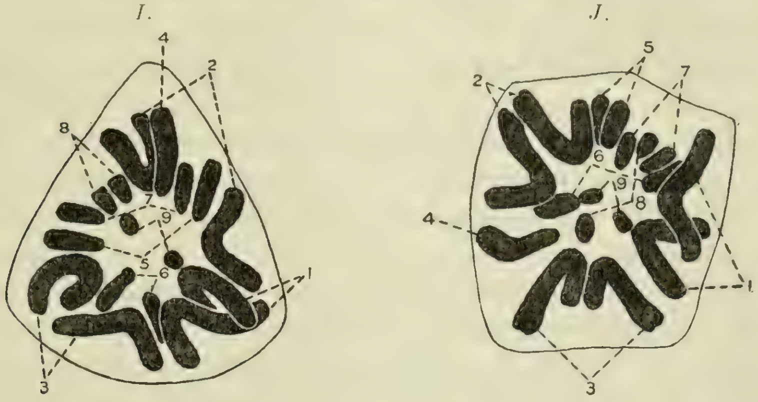
In this species the elements of the testis are smaller than in Dissosteira, but larger than in Melanoplus. The apical cell is always present at the distal end of the follicle and is surrounded by a single layer of primary spermatogonia. In the cytoplasm on one side of the nucleus is a mass of material staining deeply with Bordeaux, but it is more homogeneous and less distinctly granular than in Dissosteira. No granules staining with hematoxylin are present.

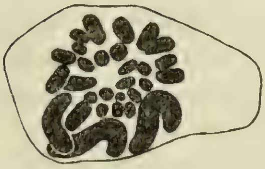
The spermatogonia are much as in Dissosteira, but stain more deeply owing to the relatively greater amount of chromatic material. The nucleus is also much more irregular in shape than in Dissosteira. This is well shown in the prophase (Plate **1**, Fig. 17, Plate **2**, Fig. 21), where the large vesicular nucleus is more easily distinguishable from the surrounding cytoplasm. The asters first appear in a deep depression on the distal side of the nucleus. At the center of each aster is a minute centrosome. In the metaphase there are seventeen chromosomes in the equatorial plate, which differ more in size than in the preceding species and thus can be more easily grouped in pairs. In Figures *I* and *J* can be seen three pairs of very large autosomes (1, 2 and 3), which differ not only in size but in shape. The chromosomes of one of the three larger pairs always have the mantle fibers attached to their middle, the two equally long arms projecting away from the spindle in the form of a letter U or letter V. In the case of the other two large pairs, the mantle fibers are attached not at the middle but nearer one end of the chromosome, the projecting arms thus being of unequal length. The next smaller chromosome (4) is the monosome. Between the monosome and the next smaller chromosomes there is a distinct break in the series. The ten smaller autosomes can easily be grouped in five pairs (5, 6, 7, 8, and 9), which constitute a nicely graded series.

Of all the species studied, Stenobothrus shows most clearly the paired relation of the autosomes. However sceptical one may be about this in the case of the other species described, there can be no doubt that in Stenobothrus there are always, with the exception of the monosome, two chromosomes of each size.

G. Steiroxys trilineata.

This is the only locustid that I have studied. As one would expect, it differs quite markedly from the Acrididae. Possibly the most striking difference is due to the fact that the cells are much smaller and richer in chromatin. In the later generations of the spermatogonia the amount of cytoplasm is very small, so that the cysts appear at first glance to be made up almost entirely of deeply staining nuclei. As in the Acrididae, there is always an apical cell (Plate **1**, Fig. 4) at the distal end of each follicle. The nucleus stains only faintly with iron hematoxylin, the network being made up largely of achromatic material, in which are imbedded scattered chromatic granules. The granular material surrounding the nucleus stains only faintly with Bordeaux.

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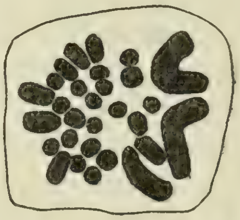
L.

FIGS. *I* and *J*. Polar views, metaphase of spermatogonia in *Stenobothrus curtipennis* showing autosome pairs. × 1450.

FIG. *K*. Polar view, metaphase of epithelial cell from vas deferens of *Steiroxys trilineata*. × 1450.

FIG. *L*. Polar view, metaphase of spermatogonium of *Steiroxys trilineata*. × 1450.

Both the primary and secondary spermatogonia in the resting stage differ greatly from the corresponding stages in the Acrididae. The nuclei are more irregular in shape and the chromatic granules are much more abundant and more evenly distributed. The monosome occupies a separate vesicle having a complete membrane of its own. It is even separated from the rest of the nucleus by a thin layer of cytoplasm. The autosomes on the contrary are all enclosed in a common membrane not being located in separate vesicles as Otte (:06) found in Locusta. The monosome usually appears as an almost homogeneous mass, but when strongly decolorized it exhibits a well defined granular structure as in the rest of the nucleus, except that the granules are much more closely aggregated. The monosome has apparently no constant position in the cell, but may occur at either pole.

During the prophase (Fig. 16) the chromosomes appear as contorted rod-shaped bodies which later shorten and thicken. In favorably situated nuclei the monosome can be distinctly seen in a separate vesicle (Fig. 16). In the equatorial plate (Fig. *L*, p. 77, and Plate **2**, Fig. 18) there are twenty-nine chromosomes, which always lie in a single plane. This species is remarkable for the ease with which the chromosomes can be counted, as they are usually so well separated that there is not the slightest difficulty in distinguishing the individual elements. There are always three chromosomes much larger than the others; two of these are V-shaped and form a symmetrical pair, while the third, a long rod-shaped element is unpaired, being, of course, the monosome. A comparison of Figures 16 and 18 shows that this is undoubtedly the chromosome which is contained in a separate vesicle during the prophase. Its relative size leaves no room for doubt on this point. The remaining twenty-six chromosomes can be readily grouped in pairs.

Figure *K* (p. 77) is a polar view of the metaphase of an epithelial cell from the vas deferens. There are probably twenty-nine chromosomes as in the spermatogonia, but they are so closely crowded that I cannot feel sure the count is correct. However, there can be no doubt that in these cells, just as in the spermatogonia, there are three chromosomes which are much larger than the rest, two of them forming a symmetrical pair, while the third is without a corresponding mate and undoubtedly represents the monosome.

3. Growth Period of the Primary Spermatocytes.

A. Dissosteira carolina.

1. *Autosomes.*

Immediately succeeding the telophase of the last spermatogonial division the chromatin becomes diffused throughout the nucleus in the form of irregular granules suspended in the linin network (Plate 2, Fig. 26). At intervals the granules may be aggregated into clumps, which are often of considerable size. This stage, which marks the beginning of the primary spermatocytes, is, I believe, comparable to the resting stage of the spermatogonia.

For the sake of convenience in describing the history of the primary spermatocytes it has seemed desirable to distinguish ten quite well marked stages, of which the condition described above is the first, or stage *a*.

During the telophase of the last spermatogonial division and the early part of stage *a* the chromatin is often shrunken away from the nuclear membrane forming a deeply staining mass eccentrically placed within the nucleus (Figs. 24, 25). Such a condition evidently corresponds to that first described by Moore ('95) to which he applied the term "synapsis." Similar appearances have been described by a number of later writers on spermatogenesis, but, usually, there seems to be more or less uncertainty as to whether this condition is normal. At first I was inclined to believe that this is a normal stage which lasts but a very short time, since cells in this condition occur quite commonly during the late telophase and beginning of stage *a*, but at no other time. However, a more careful study has convinced me that such a condition of the chromatin is, in reality, an artifact. I am led to this conclusion more especially by the fact that in almost every case the cells of the peripheral layer, which are in contact with the follicular wall and thus in the most favorable position to be acted on promptly by the fixing agent, show no such contraction of the chromatin. It is, however, undoubtedly true that at this stage, and at no other, the chromatin shows a marked tendency to contract away from the nuclear membrane if not properly fixed.

Stage *a* is quickly followed by stage *b* (Fig. 28), which is characterized by the collection of the chromatin into more or less definite, elongated masses. These masses are connected with one another and with the nuclear membrane by fine linin fibrils and have, in general, the same orientation as the chromosomes of the preceding telophase. They appear to be composed of a very much contorted thread made up of chromatin granules imbedded in a linin matrix. This thread is so contorted and bent upon itself that it is impossible in this species to determine whether each mass is composed of one or several distinct threads. The structure of these masses can be made out much better in Chortophaga viridifasciata, in the account of which they will be described in detail. It is sufficient here to state that they are much more distinct and widely separated in this species than in any other, while their number is approximately the same as that of the spermatogonial chromosomes. We may conclude, then, that each mass of chromatin is a univalent autosome, which in Chortophaga can be seen to be transformed, by a sort of unraveling process, into a chromatin thread. In Dissosteira it is impossible to make out satisfactorily the formation of the threads, but in the next stage (*c*) the nucleus contains probably several much convoluted threads (Figs. 29, 30) composed of linin, in which small rounded chromatin granules are imbedded at short, and fairly regular intervals. Extending out from each thread, and connecting it with adjoining threads, are fine linin fibrils. It is impossible at this stage to determine by observation whether there is one or a large number of chromatin threads, but, for reasons given above, I believe that there are really a number of distinct threads, and that there is at no time a continuous spireme. During this stage one or two small plasmosomes, staining deeply with Bordeaux, appear in the nucleus.

The next stage (*d*) is characterized by polarity in the arrangement of the spireme. In favorable cases it can be plainly seen that the threads are now attached at one side to the nuclear membrane (Figs. 31, 32). Probably the attachment takes place somewhat earlier, but is not distinguishable on account of the more tortuous course taken by the spireme threads. When this polarity of the spireme first becomes evident the threads still take such a tortuous course through the nucleus that the polarity is distinguishable only near the region where the threads are attached to the nuclear membrane. However, during the latter part of this stage the polar arrangement of the threads becomes much more distinct; but it is never as marked in this species as in some other Orthoptera. A careful study of the nucleus at this stage shows that the spireme is really in the form of loops attached by their free ends to the nuclear membrane at a common point. At first glance the number of loops at this time appears to be considerably greater than at a somewhat later stage. However, I believe this appearance to be deceptive, and due to the fact that the loops at this stage have a great length, which, in some cases at least, is probably several times the diameter of the nucleus. This results in their being more or less bent upon themselves, so that a single loop may extend across the nucleus several times. During the latter part of this stage the loops become much shorter, while at the same time the nucleus increases markedly in size, so that in most cases the arms of the loops are not much longer than the diameter of the nucleus.

I have been unable to count at this stage the loops in Dissosteira but in Hippiscus, where the material is more favorable, the number seems to be about one-half that of the spermatogonial autosomes. It is impossible to count the loops with perfect accuracy, but I am convinced that the number is much less than that of the autosomes in the spermatogonia. It thus appears probable that each loop represents two univalent autosomes of the spermatogonia which have become joined end to end. This interpretation is strongly supported by stage *b*, where, as we have seen, there is good reason to believe that each autosome is converted into a *single* chromatin thread. If, then, we imagine these threads to become united in pairs at one end, while the other end becomes attached to the nuclear membrane, we should have the arrangement of threads seen in stage *d*. This I believe is the manner in which the loops are formed, although it is obviously impossible to demonstrate it, since in the earlier stages the threads are so closely crowded and pursue such tortuous courses that it is impossible to follow one for any great distance. The structure of the spireme-thread during stage *d* is practically the same as in the preceding stage, being made up of a single row of chromatin granules imbedded in a linin thread.

The loops of the spireme are usually attached to the nuclear membrane at a point adjacent to the greater mass of cytoplasm and to the interzonal body. In other words, the attachment is at the distal pole of the nucleus, i. e. at the pole nearest the plane of the last division. However, in some cases the point of attachment may be at first as much as 90°, or even more, from the distal pole of the cell, but during the later stages of the growth period it is almost invariably at this pole. Obviously, this implies in some cases a considerable revolution of the nucleus, but I do not believe such a revolution can be considered general, since in the majority of spermatocytes the loops are attached at the distal pole when this polar arrangement first becomes evident. It is only fair to say, however, that at this stage it is often difficult to distinguish the distal pole on account of the small amount of cytoplasm.

Instead of considering that we have here a regular revolution of the nucleus, it seems to me more reasonable to suppose the variation in the position of the point of attachment in the earlier stages to be due to the mutual pressure of surrounding cells. The interzonal body, which is the criterion used to distinguish the distal pole, invariably lies in the part of the cell containing the greatest amount of cytoplasm, and the shape of any cell is certainly largely determined by the pressure of surrounding cells. It would, therefore, seem only reasonable to suppose that the relative positions of the nucleus and interzonal body might be considerably influenced in this way. The revolution of the nucleus in some cases would, then, be simply a readjustment in the cell to a more or less definitely fixed polarity which had been temporarily lost.

In the following stage (*e*) the chromatin granules (Plate 2, Fig. 34), which are distributed along the spireme threads, divide so that each loop shows more or less distinctly a longitudinal split. It should be clearly understood that this splitting does not extend to the linin, which now forms a flattened ribbon like structure, in which the paired chromatin granules are imbedded. This stage undoubtedly corresponds to the split spireme of authors. I believe this double series of chromatin granules to be formed by the splitting of the single series of granules of the preceding stage, rather than by a side to side union of two distinct threads, as has been held by a number of recent writers to be the case in other forms. Long and careful study of a large number of cells in this and preceding stages has convinced me that there is little evidence in this species (or in other Orthoptera) of a side to side union of the spireme-threads. Occasionally during the preceding stage two threads can be seen lying parallel for a short distance, but if carefully followed they can in almost all cases be seen to diverge again. This, combined with the fact that such structures are comparatively rare, has convinced me that they are only accidental. Moreover, in stage *e* part of the chromatin granules along a given thread may be single while others are double. The single granules are usually elongated transversely to the thread as though preparing to divide into two. Of course it may be argued that these granules are formed by the fusion of two originally distinct granules, but there is little to support such an interpretation, since there is no evidence that the longitudinal split later becomes obliterated by the fusion of the halves. However, the whole question will be taken up later, in a discussion of the literature, where it can be considered to greater advantage. During stage *e* the polar arrangement of the spireme is more marked than at any other time. Figure 33 shows a spireme as seen from the pole. It is evident that the loops all tend to converge at a single point, where they are attached to the nuclear membrane. In reality, the ends of all the loops do not come together at a common point, but some unite with each other a short distance from the point of attachment, so that the threads often appear to branch. It is almost impossible to make out the exact arrangement of the threads near the point of attachment, since they are so closely crowded together (often overlying one another), and also since in this region the chromatin granules are usually much larger, and distributed along the linin thread at much longer intervals than elsewhere. In such cases it is often very difficult to follow the lightly staining linin thread.

The next stage (*f*) marks the end of the growth period (Plate 3, Fig. 46). The spermatocytes have now reached their greatest size, both as regards the nucleus and the cytoplasm, while the chromatin has become more diffuse than at any other stage. Judging from the number of cells found in this condition, this is the longest of the spermatocyte stages.

The transition from stage *e* to stage *f* is gradual and consists chiefly of an opening out of the loops, so that most of them come to lie close under the nuclear membrane. The result is that in most cases the polarity of the spireme seems to have been lost and the whole appearance suggests that there is a continuous spireme extending around the periphery of the nucleus. Farmer and Moore (:05) have, indeed, held that such is the case in Periplaneta, but in the Orthoptera which I have examined I do not believe there is a continuous spireme at any time. In fact, during stage *f*, when in most cases there appears to be no trace of the former polarity, the loops for a considerable time at least retain their original attachment to the nuclear membrane. In nuclei which are favorably oriented so as to afford a view of the distal pole, the ends of the loops can still be seen to be attached to the nuclear membrane as in the preceding stages, but owing to the tortuous course of the threads around the periphery of the nucleus, all trace of the radiate arrangement is lost except in the immediate vicinity of the distal pole. In some species of Acrididae, where the polarity is much more marked, it is plainly distinguishable even at this stage.

Concurrently with the opening out of the spireme loops the longitudinal split becomes indistinct, and the thread seems to stain much less deeply. This appears to be due chiefly to the chromatin granules becoming broken up into much finer particles, which are irregularly distributed along the linin thread. At the same time the amount of linin is considerably increased, and the fine radiating fibrils which extend out through the nucleus become more abundant.

During stage *f* the plasmosomes, of which there are usually two or three, reach their greatest size, and in the latter part of this, or in the following stage become more or less irregular in shape and often break up into a number of irregular fragments. At the same time they tend to stain with iron hematoxylin, so that they often resemble masses of chromatin.

I have often noticed a rounded body in the cytoplasm which strikingly resembles a plasmosome. This body is more common during stage *f*, although I have occasionally found it at a considerably earlier period. It is often closely applied to the nuclear membrane, but whether it is a plasmosome which has been extruded from the nucleus, I have been unable to determine. Certainly, some of the plasmosomes are never cast out into the cytoplasm, but degenerate within the nucleus at a little later stage.

2. *Monosome.*

Having described the changes in the autosomes during the growth period, I will now take up the history of the monosome during the same period. During the telophase of the last spermatogonial division (Plate **2**, Figs. 22, 23) the monosome retains its compact structure and remains enclosed in a separate vesicle. At this stage a deep furrow or groove can often be distinguished extending along one side of the nucleus, and in such cases the monosome usually lies along the opening of the groove. This is especially well shown in cases where, on account of imperfect fixation, the chromatin is shrunken away from the nuclear membrane (Figs. 24, 25). Very often the monosome is more or less completely divided by a transverse constriction into two unequal parts.

During the succeeding resting stage (*a*) of the primary spermatocyte the monosome still remains enclosed in a separate vesicle, and, as in the preceding stages, is an elongated homogeneous deeply staining element often showing a more or less distinct bipartite structure (Fig. 23). In stage *b* (Fig. 28, Plate **7**, Figs. 99, 100) there is little change in the appearance of the monosome, but in the next stage (*c*) it no longer lies in a distinct vesicle, but comes to lie within the common nuclear membrane (Fig. 29). During this stage there is little change in the monosome except that it becomes somewhat flattened and irregular in shape.

In stage *d* the monosome (Plate **2**, Figs. 31, 32; Plate **7**, Figs. 101,

102) forms an irregular flattened plate closely apposed to the nuclear membrane. It is usually somewhat vacuolated and tapers at one end, which is attached to the nucleus at the same point as the autosomes. The monosome usually lies near the distal pole of the nucleus rarely being distant more than 90° from it. During the following stage (*e*) there is very little change (Fig. 34), but during stage *f* the monosome undergoes a very interesting metamorphosis. At the beginning of this stage the monosome forms an irregular flattened plate closely applied to the nuclear membrane, and connected at one end with the distal pole of the nucleus. A view of the monosome *en face* (Plate **7**, Figs. 103, 104) shows that it does not stain uniformly, but contains several lighter areas, where it is probably thinner than elsewhere. When strongly decolorized it can be seen to be composed of minute deeply staining granules imbedded in a less deeply staining matrix. At a little later stage (Figs. 105–107) the thin areas have become broken through so that the monosome becomes converted into a loop both ends of which are attached to the nuclear membrane at the distal pole. At this time the monosome has a very ragged outline and stains but little deeper than the other nuclear elements, so that it is often difficult to distinguish it.

During the entire growth period the mitochondrion can be distinguished as irregular masses scattered through the cytoplasm and staining with Bordeaux more deeply than the latter. Usually, it is especially abundant at the distal pole of the cell, but also forms an irregular layer around the nucleus. At first the amount of mitochondrion is small, but it increases rapidly during the growth period and becomes a conspicuous element of the cytoplasm. In material fixed with Flemming's fluid the mitochondrion is much more conspicuous and stains much more deeply than in Hermann material. In Flemming material it is usually distinctly granular and in some cases fibrillar.

The interzonal body, which lies at the distal pole, so closely resembles the mitochondrion that it is often impossible to distinguish one from the other. In most cases the interzonal body can be identified with certainty only where it can be traced into an adjoining cell.

B. Arphia tenebrosa.

1. *Monosome.*

The spermatocytes in this species are very similar to those in Dissosteira, but are in some respects even more favorable for study. Plate

2, Figure 35 shows the split spireme (stage *e*) from the distal pole. The monosome, forming a flattened plate composed of deeply staining granules imbedded in a lighter matrix, lies close to the pole (on the lower side in the figure) and is connected with it by a flattened process, which occupies most of the polar area. Figures 112 and 114 (Plate **7**) represent variations in the monosome at practically this stage. In Figure 112 the monosome is attached to the pole by two filaments, while in Figure 114 it lies almost directly at the pole. Figure 113 shows a slightly later stage, in which the monosome has become converted into a loop, but still retains its connection with the pole.

In one individual a very interesting abnormality was found, all the germ cells of the testis showing constantly two monosomes. Unfortunately I have been able to find in this individual only one dividing spermatogonium in which the chromosomes could be counted with any accuracy. This cell contained twenty-four chromosomes instead of the normal number, twenty-three. In all stages of the spermatocytes two well defined monosomes could be distinguished. Figure 27 (Plate **2**) shows a primary spermatocyte during stage *a* in which both monosomes are easily distinguishable. On account of the very thick sections in this series I was unable to determine whether the two monosomes are enclosed within separate vesicles. Figure 37 (Plate **3**) represents a later stage (*d*) from the same individual, the two monosomes being clearly shown. Apparently, both monosomes independently pass through the same stages as the single monosome of normal individuals. There is, however, a slight difference, in that one monosome shows a greater tendency to disintegrate than the other.

C. Hippiscus tuberculatus.

1. *Autosomes.*

The growth period in Hippiscus closely resembles that already described in Dissosteira. However, on account of the chromatin granules being located at greater intervals along the spireme thread, the formation of the split spireme can be studied here to greater advantage. Figures 38–40 (Plate **3**) show a few of the spireme threads at successively later stages. During the early part of stage *d* (Fig. 38) the spireme is composed of a single row of chromatin granules imbedded in a linin thread. At a little later stage (Fig. 39) the granules are clearly larger and usually elongated transversely to the linin thread. At the left in Figure 39 is shown a portion of a thread in which the granules show evident signs of division. The spireme at this time is flattened and where the narrow side is turned toward the observer, as in a portion of the thread at the right in the figure, it appears practically the same as at an earlier stage (cf. Fig. 38). Later (stage *e*, Fig. 40) the chromatin granules are clearly in pairs and the spireme ribbon has become still wider. However, when the ribbon is seen edgewise it still appears to contain only a single row of granules. The appearance of the spireme during this period lends little support to the view that the double threads shown in Figure 40 are formed by an approximation in pairs of the separate threads shown in Figure 38. As in Dissosteira, occasionally two single threads can be seen lying side by side for some distance, but such an arrangement appears to be purely accidental. On the supposition that the double threads are formed by the approximation of two single threads, it is impossible to explain such stages as are shown in Figure 39, which are common and are certainly intermediate between those of Figures 38 and 40. The position of such spermatocytes in the follicle leaves no room for doubt on this point. Furthermore, I have carefully counted the number of threads connected with the distal pole during stage *d*, before there is any trace of the longitudinal split, and although it is impossible to obtain an accurate count of the threads at this time, yet it appears to be approximately the same as the number of autosomes in the spermatogonia. In other words, the number of spireme loops is approximately one-half the number of spermatogonial autosomes. If the double spireme is formed by a side-by-side conjugation of single threads, there should be at this time as many loops as autosomes in the spermatogonia.

2. *Monosome.*

The monosome during the growth period differs from the same element in Dissosteira only in small and unimportant details.

D. Chortophaga viridifasciata.

1. *Autosomes.*

The history of the growth period in this species presents no essential differences from that described for Dissosteira, except in the case of stage *b*. During this stage the chromatin collects into much more distinct and widely separated masses than in Dissosteira, so that the true significance of this stage can here be followed to much better advantage. Figures 41 and 42 (Plate 3) show two cells in this stage cut at right angles to each other. The chromatin masses are here plainly distinguishable from one another, and each appears to be composed of a much convoluted and tangled chromatin thread, which is bound into a more or less compact mass by linin fibrils. Although, it is impossible to count these masses accurately owing to the great difference in their size and to the fact that a cross section of all cannot be obtained in the same plane, their number is undoubtedly approximately that of the autosomes of the spermatogonia. Moreover, the masses of chromatin have approximately the same orientation that the autosomes had during the preceding telophase of the last spermatogonial division. For these reasons I believe we are justified in concluding that each of these masses represents a univalent autosome. Later, each mass becomes converted into a *single* chromatin thread by a sort of unraveling process. In Figures 43 and 44 (Plate **3**) four of these masses are drawn in detail. This is a slightly later stage than that shown in Figure 41, and the method of formation of the chromatin thread is well shown. If each of these masses represents, as I believe, a univalent chromosome, then it is evident that during this stage each chromosome becomes converted into a long thread composed of a single row of chromatin granules imbedded in a linin matrix.

In the following stage (*c*) these threads become evenly distributed through the nucleus, giving the appearance of a very tortuous but continuous spireme, and they have been so interpreted by most investigators. There is, however, no evidence that such is the case. In the next stage, which rapidly follows this, the spireme, just as in Dissosteira, is made up of loops the ends of which are attached to the nuclear membrane at the distal pole. Since the number of loops even at this early stage is, I believe, only one-half that of the autosomes in the spermatogonia, it is probable that during stage *c*, or earlier, the chromatin threads unite in pairs end to end to form loops, while the opposite ends become attached to the nuclear membrane at its distal pole. However, it is impossible to say precisely when the conjugation of the autosomes takes place. Possibly it may occur as early as stage *b*, as I have in a few cases seen structures which might be interpreted as the result of the fusion of two chromatin masses end to end; but as such structures may be only accidental, I have not thought it best to attach any weight to them.

2. *Monosome.*

The history of the monosome during the growth period is practically the same as in Dissosteria.

E. Melanoplus femoratus.

1. *Autosomes.*

During the growth period the same stages can be recognized as in Dissosteira. In stage *b* (Plate **3**, Fig. 45) the chromatin is aggregated into well defined masses, as in Chortophaga, and the spireme threads are later formed from these masses in a similar manner.

2. *Monosome.*

The changes which take place in the monosome during this period are very different from anything occurring in the preceding species, but somewhat similar structures are found in Stenobothrus. However, on account of the small size of the monosome in Melanoplus, it has been impossible to trace its development as completely as in that species During the early growth period the monosome forms a deeply staining, somewhat flattened element apposed to the nuclear membrane. In stage *d* it usually shows a distinctly bipartite structure (Plate **7**, Figs. 115, 116), one component being slightly larger than the other. At this time the two components are usually close together, but occasionally they may be some distance apart. Later (stage *e*) the monosome becomes distinctly vacuolated and one of the components begins to lengthen (Figs. 117, 118); it continues to do so until, in the following stage (*f*), it forms a much elongated granular body, which closely resembles one of the spireme threads, and is on that account often difficult to distinguish. The other component retains its compact form.

The mitochondrion is relatively more abundant than in Dissosteira and is scattered irregularly through the cytoplasm forming small, finely granular, flocculent masses, which stain deeply with Bordeaux.

F. Stenobothrus curtipennis.

1. *Autosomes.*

The growth period in this species is, in general, much as in Dissosteira, although, on account of the relatively much greater amount of chromatin, the material is, in many respects, less favorable for study. Figure 47 (Plate **3**) shows the first stage (*a*) of the spermatocytes. Figure 48 shows the split spireme stage (*e*). Owing to the large size of the loops, the polarity is never as marked as in the preceding species, but the longitudinal split often shows with almost diagrammatic clearness. The chromatin granules are large, and can be clearly seen arranged in pairs at nearly regular intervals along the linin thread. Figures 49 and 50 show the spireme as seen from the distal pole, while Figure 51 shows a cross section of the loops just below the point where they are attached to the nuclear membrane. In addition to the monosome, the cut ends of fifteen or sixteen threads can be distinguished in the section, which is the number we should expect to find, since there are sixteen autosomes in the spermatogonia. In stage *f* (Fig. 52) the loops open out as in the preceding species and tend to take a peripheral position. The chromatin granules break up into still finer particles, and the longitudinal split becomes less evident. However, in this species there is no such obliteration of the longitudinal split as in Dissosteira, but throughout stage *f* it remains fairly distinct.

2. *Monosome.*

The history of the monosome during the growth period is quite different from that in Dissosteira, but shows many points of resemblance to Melanoplus. However, owing to its greater size the changes in the monosome can be followed to much better advantage in this species. During stage *a* (Fig. 47) the monosome is an elongated, deeply staining, homogeneous body enclosed in a separate vesicle. Rarely, it is divided by a transverse constriction into two nearly equal parts. In stage *b* there is little change in the appearance of the monosome, but, as in the preceding species, it becomes enclosed within the common nuclear membrane. In the following stage (*c*) (Plate **7**, Figs. 125–128) the monosome forms a more or less flattened plate, in which several small vacuoles can usually be distinguished. A little later (stage *d*) it begins to elongate (Figs. 129, 130) and a process grows out from one end. In the following stage (*e*) this process may reach a considerable length (Figs. 131–132). During this stage the bipartite structure of the monosome becomes evident. One part is a flattened, rounded or somewhat elongated body with smooth contours, and contains numerous small vacuoles. The other part is of nearly the same size but greatly elongated and distinctly granular. In fact it strikingly resembles the spireme, except that the chromatin granules are larger and crowded together more closely. When the monosome lies at some distance from the distal pole it is connected with it by means of this elongated part (Plate **3**, Fig. 49; Plate **7**, Fig. 131). However, if located near this pole, as is usually the case, the elongated part extends out free into the nucleus (Fig. 132). In this and succeeding stages there is almost invariably a plasmosome in close proximity to the monosome, usually in immediate contact with it, though in exceptional cases at some distance from it.

During stage *f* the bipartite structure of the monosome is very marked (Plate **3**, Fig. 52; Plate **7**, Figs. 133–138). One part, as in the preceding stage, can be distinguished by its more compact structure and smooth contours; the other or elongated part is distinctly granular with rough, ragged contours. As shown in the figures, it is usually curved, and sometimes the two ends may come together so as to form a ring (Fig. 133). In some cases this ring may entirely surround the non-granular part, while the two parts are more or less intimately connected.

The changes in the cytoplasm during the growth period are much the same as those described in Dissosteira.

G. Steiroxys trilineata.

1. *Autosomes.*

In this locustid the spermatocytes pass through essentially the same stages as in the Acrididae, but owing to the small size of the cells and relatively great amount of chromatin the material is much less favorable for study. Figure 54 (Plate **3**) shows the first stage (*a*) of the primary spermatocyte, while Figure 53 is an imperfectly fixed spermatocyte in the same stage. Owing to the chromatin being shrunken away from the nuclear wall the concavity on one side of the nucleus shows much better than on well fixed material. Figure 55 is that of a somewhat later stage (*c*), in which, as in the Acrididae, fine chromatin granules are distributed along a much convoluted spireme. In this species, however, the chromatin is in much finer granules and more irregularly distributed along the thread than in any of the Acrididae. In the next stage (*d*) the spireme has the usual polar arrangement (Fig. 56) and at a little later stage (*e*) shows (Plate **5**, Fig. 69) a more or less distinct longitudinal splitting. This splitting is never as marked as in many of the Acrididae, but I believe there can be no doubt that such a stage occurs. I have failed to find any evidence of the side-to-side union of the spireme threads which Otte (:**06**) has found in Locusta. During stage *f* all traces of the longitudinal splitting disappear owing to the fact that the chromatin granules become broken up into finer particles and are irregularly distributed along the spireme thread, which at this time has a considerable size. Throughout this stage the spireme retains its distinct polarity, although, as in the Acrididae, the loops tend to assume a peripheral position.

2. *Monosome.*

The monosome during the early part of the growth period (stage *a*) is inclosed in a separate vesicle lying next the concave side of the nucleus, but always separated from the nuclear wall by a thin layer of cytoplasm (Plate **3**, Figs. 53, 54). At this time the monosome usually appears as a rounded, deeply staining, homogeneous mass, but on strong decolorizing shows a distinct granular structure. The whole appearance is strikingly like that of the same element in the resting spermatogonium. Later the monosome becomes flattened, so that it has the shape of a plano-convex lens, the flat side being applied to the exterior of the nuclear membrane (Fig. 55). In the following stage (*d*) the monosome (Fig. 56; Plate **7**, Fig. 151) has become enclosed within the nuclear membrane. It now forms a deeply staining, somewhat elongated and flattened element closely applied to the nuclear wall and connected at one end with the distal pole; but it soon becomes converted into a U-shaped body by the development of a longitudinal split (Figs. 152, 153). In stage *f* the monosome (Fig. 154) forms an irregular, vacuolated plate, the arms of the U having fused along their entire length.

In the spermatocytes of Steiroxys the mitochondrion has a very different form from that found in the Acrididae. Instead of being distributed irregularly through the cell, it is collected into a rounded mass, which lies in the cytoplasm at the distal pole (Plate **3**, Fig. 56). This body, which is distinguished from the surrounding cytoplasm by its deeper stain, is very finely granular and usually shows a still more deeply staining layer around the periphery. Sometimes the entire body is composed of several deeply staining rings separated by lighter areas. A very similar condition of the mitochondrion has been described by Otte (:**06**) in Locusta. In addition to the mitochondrion an interzonal body is present at the distal pole and appears essentially the same as in Dissosteira.

4. The Maturation Period.

A. Dissosteira carolina.

1. *Autosomes.*

At the end of the growth period the loops of the spireme, though still showing their polar arrangement, have taken a peripheral position, while the longitudinal split has become indistinct. In the next stage (*g*), which marks the beginning of the maturation period (Plate **4**, Figs. 57, 58), the spireme loops have become detached from the nuclear membrane and are irregularly distributed through the nucleus. The chromatin becomes aggregated into larger granules, while the longitudinal split is more distinct than at any previous stage, the space between the two series of chromatin granules being wider than at any other time.

In stage *h* (Fig. 59) the loops have become converted into definitive tetrads. These vary greatly in shape, but may be roughly grouped into three types: (1) straight or curved rods, (2) crosses, and (3) rings, or loops with their free ends crossed. These types are all, I believe, modifications of a common fundamental form composed of two longitudinally split threads or rods of equal length joined end to end. Each tetrad is apparently formed from a single loop of the spireme in the following manner: When the loops first become free they are apparently of uniform appearance throughout their entire length, but a little later the longitudinal split begins to widen at the middle of the loop forming a diamond shaped opening (Plate **7**, Figs. 160, 161). This opening I believe to be at the point of union of two univalent autosomes of the spermatogonia. These rod-shaped autosomes are usually more or less curved, rarely straight. This is the simplest type of tetrad which I have found in this species, but in Hippiscus a still simpler form occurs (Fig. 159). In this genus there is no widening of the longitudinal split at the middle, but there is at this point a distinct break in the chromatin, the intervening space being bridged over by linin.

The second type of tetrad has the form of a cross (Figs. 165–167) in which the four arms may be approximately equal in length, or one pair may be longer than the other. A further complication is caused by the arms being usually more or less bent, the free ends of each pair tending to approach each other. However, both pairs of arms never bend in the same direction. Figure 169 shows a case where this process has been carried to an extreme. This form of tetrad is, like the former type, derived from two longitudinally split rods, and in the same way, except that here the process is carried much farther. The halves of each rod diverge at the point of union of the univalent autosomes along a line at right angles to the longitudinal split, while at the same time the free ends of each pair of arms bend toward each other. Obviously, in this type, where the two pairs of arms may be of nearly equal length, it is often impossible to recognize the point of union of the univalent components, whether at the ends of one or the other pair of arms.

The third type of tetrad may be looked on as a still further modification of the common fundamental form. In this type the halves of the split rods do not diverge along the line of union of the univalent components to as great an extent as in the preceding type, so that the arms developed along this line are always shorter than the others. By the bending of the two longer arms their tips approach each other until the arms meet or even cross each other. This gives rise to closed rings or crossed loops (Figs. 170–178). The latter when viewed in a certain direction may appear x-shaped. The free ends of the crossed loops may unite giving rise to a figure of 8. Extending out from the tetrads in all directions are fine linin fibrils of varying lengths, which give to the tetrads a characteristic hairy appearance. The longer fibrils are continuous with a well defined linin network which extends throughout the nucleus.

The later changes in the structures of the tetrads appear to be due entirely to a concentration of the chromatin.

In stage *i* (Plate 4, Fig. 60) the chromatin becomes much more compact, while the longitudinal split is much less distinct, and at the end of this stage (Fig. 61) has become almost entirely obliterated. The tetrads for the most part are arranged around the periphery of the nucleus in contact with the nuclear wall. As in the preceding stage, there are large numbers of linin fibrils extending out from the tetrads in all directions. The linin network is more highly developed than at any previous period; so much so, in fact, that the nucleus often appears nearly as dense as the surrounding cytoplasm.

The mitochondrion is more abundant at this stage than at any other time and forms irregular, very finely granular masses which are distributed around the nucleus at irregular intervals and stain with Bordeaux.

At this time two minute centrosomes surrounded by well developed asters are seen in the cytoplasm close to the nuclear membrane (Fig.

61). Apparently the centrosomes are already some distance apart when first distinguishable, and continue to migrate around the nucleus until they come to lie opposite each other. The nuclear membrane then gradually breaks down, first disappearing in the vicinity of the asters. As the nuclear membrane disappears the astral rays extend into the nucleus and become attached to the chromosomes. Figure 62 shows a somewhat later stage, after the spindle is fully formed, but before the chromosomes are drawn into the equatorial plane. The autosomes have now become homogeneous and have perfectly smooth contours, but they retain, in general, the characteristic forms seen in the earlier stages. Figure 63 shows the metaphase of the first maturation division. As the chromosomes become drawn into the equatorial plane, the rod-shaped forms are arranged with their long axes parallel to the axis of the spindle, the mantle fibers being attached at each end (Plate **7**, Fig. 164). Thus the spindle fibers from one pole are attached to one of the univalent components of the bivalent autosomes, those from the other pole to the other univalent component. The cross-shaped autosomes become so arranged that one pair of arms lies along the spindle while the arms of the other pair project away from the axis of the spindle and, at the same time, make a considerable angle with each other, so that in a polar view they appear V-shaped, the apex of the V being directed toward the spindle. In these autosomes it is impossible to determine whether the plane of union of the univalent components is at right angles to the axis of the spindle or parallel to it, but from analogy with the other types it is probably at right angles. In the case of the closed rings and crossed loops the evidence is more satisfactory. These chromosomes become so arranged on the spindle that the free end of each univalent component is attached to mantle fibers from the *opposite* pole of the spindle, while the apex, which represents the point of union of these components, projects away from the spindle (compare Figs. 179, 180).

In the metaphase of the first maturation division there are always twelve chromosomes, eleven of them being bivalent autosomes, which vary greatly in size. Figure *N* is a polar view of the equatorial plate in two adjoining cells and shows that in general the autosomes have the same size relations as the autosome pairs of the spermatogonia, although in the spermatocytes the relative size of the chromosomes, on account of their irregular form, is not as clearly marked. Inasmuch as the univalent components of each bivalent autosome are of equal size, there seems to be no good reason for doubting that each bivalent autosome is formed by the union of two homologous autosomes of the spermatogonia.

An extended study of the chromosomes during the maturation period has convinced me that every spermatocyte possesses the same number of bivalent autosomes of each of the three types. This is not a statement which can be easily demonstrated, since only rarely can the structure of all the elements of any one cell be determined with certainty. Viewed at slightly different angles the appearance of a chromosome may vary greatly, and at certain angles a given chromosome may be indistinguishable from one of a different type. Polar views of the equatorial plate are most favorable for identifying the different forms, but even in such cases it is often impossible to be sure of the structure of all the elements. However, in the most favorable

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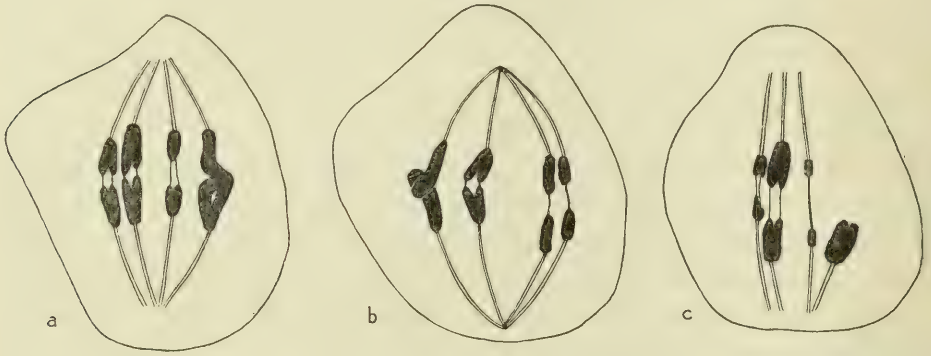
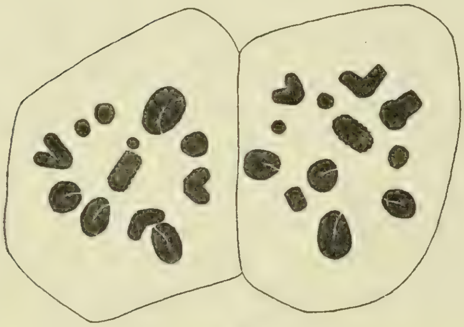
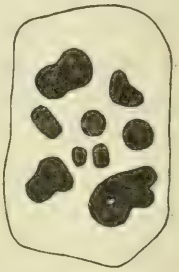




N.



NN.

Figs. *Ma, Mb, Mc.* Successive sections of spermatocyte of *Dissosteira carolina* during the early anaphase of the first division. All the chromosomes are shown, the monosome being at the right in Fig. *Mc.* × 966.

Fig. *N.*— Polar view, metaphase of first maturation division of *Dissosteira carolina.* × 966.

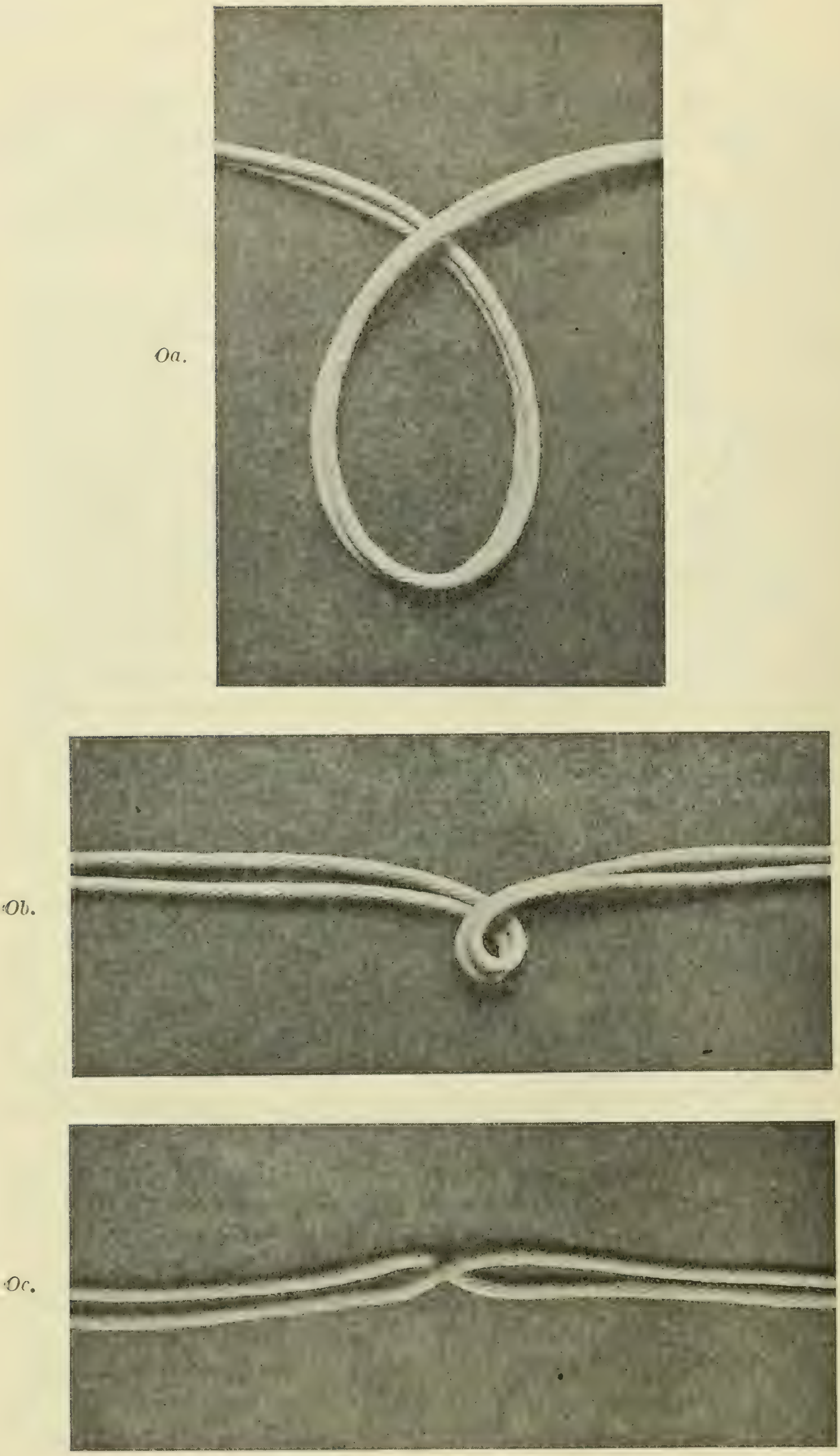
Fig. *NN.*— Polar view, metaphase of first maturation division of *Stenobothrus curtipennis.* × 966.

cases it is usually possible to recognize three autosomes of the rod type, two of which are much smaller than the other chromosomes, while the third is medium sized; three autosomes of the cross type, of which two are medium sized and the third is one of the larger chromosomes; and five autosomes of the ring and crossed-loop type, of which four are large and one medium sized.

As the chromosomes divide, the rod-shaped forms first become dumbbell-shaped by the formation of a transverse constriction, which continues to deepen until the two components are entirely separated. The cross-shaped chromosomes first become converted into rods by an elongation in the direction of the spindle axis and a corresponding shortening of the transverse arms. Later they become dumbbell-shaped and finally divide as in the former case. In the rings and crossed loops the process is more complicated. The ends of the univalent components are attached to mantle fibers connected with the more distant pole. Consequently as the chromosome divides its univalent components are pulled past each other (Plate **4**, Figs. 63, 65, Plate **7**, Figs. 179, 180). In all cases as the univalent components separate the longitudinal split, which has been temporarily hidden, again appears so that the chromosomes as they move toward the pole are composed of *two* rods, which may lie parallel or may diverge at the ends nearest the equator of the spindle, giving rise to the well known V-shaped chromosomes (Fig. 65, Fig. *M*) so characteristic of the heterotypical mitosis. As the ring- or loop-shaped autosomes (Figs. 181, 182) are pulled out during division and the longitudinal split reappears, it can be plainly seen in favorable cases that the rods resulting from the longitudinal split do not lie parallel but are crossed at the middle of the dividing autosome. Later the autosomes divide at this point, which, as the slight enlargement indicates, is the place where the univalent components are joined. I regard this crossed condition of the two longitudinal halves of the autosomes as strong confirmatory evidence of my interpretation of their structure.

If the reader will take two flexible, parallel wires, or stiff cords, and bend them until the ends are crossed (Fig. *Oa*), thus forming a loop, as in the case of the tetrads, he can get a much clearer idea of the method of separation than by any description. Now, by pulling the two ends in opposite directions the loop will diminish in size until a kink (Fig. *Ob*) develops in the middle, and when the wires are fully straightened out (Fig. *Oc*) they will be crossed just as the autosomes are in Figures 181 and 182.

As a result of their arrangement on the spindle, the univalent com-



FIGS. *Oa*, *Ob*, *Oc*.— Photographs of models to illustrate the method of division of tetrads.

ponent of each bivalent autosome must therefore separate from its mate during the first maturation division. In the case of the cross-shaped type it is impossible to determine whether or not the univalent components separate from each other during this division, but it seems as though the division in these forms was the same as in the others.

The first maturation division is, then, a true reduction division in Weismann's sense of the term, since it separates the univalent components of the bivalent autosomes.

Figure 65 (Plate **4**) shows the beginning of the anaphase (compare also Figure *Ma-Mc,* p. 96) and Figure 66, the late anaphase of the first division. At the end of the anaphase the chromosomes are collected at the ends of the spindle into two daughter groups, in which the outlines of the elements can be recognized only with difficulty. The centrosomes, which, as in the spermatogonial divisions, reach their greatest size during the metaphase, have entirely disappeared at this stage. The interzonal filaments form a deeply staining fibrillar mass around the periphery of the region formerly occupied by the spindle. This mass stains much more deeply in a region midway between the chromatin masses than elsewhere. The explanation, I believe, is that in this region part at least of the mitochondrion, which was collected around the middle of the spindle during the preceding stages, has become mingled with the interzonal filaments. Later, as the constriction between the two daughter cells deepens, the interzonal filaments are forced together and finally disintegrate.

Figure 75 (Plate **5**) shows a secondary spermatocyte in the "semi-resting" stage. A nuclear membrane is formed and the autosomes have become partially broken down, but show a more or less distinct V-shape, the space between the two arms being the longitudinal split. Judging from the scarcity of cells in this stage it is of short duration. Figure 76 shows the prophase and Figure 77 the metaphase of the second maturation division. The more or less curved rod-shaped chromosomes are arranged in pairs around the periphery of the spindle (Figs. *Q*, *R*). Each pair represents a V-shaped daughter chromosome of the first division. The chromosomes show practically the same size relations as the chromosome pairs of the spermatogonia, but on account of their more irregular shape the difference in size is not as evident. During the anaphase of the second division (Figs. 78, 79) one member of each pair passes to each pole of the spindle. Figure 80 shows a telophase of the second division.

Since, during the second maturation division, the autosomes divide along the plane of the longitudinal split which first appeared in the polar spireme, this division is equational.

2. *Monosome.*

Having followed the history of the autosomes during the maturation period, we will now trace the monosome through the same period. At the beginning of the maturation period the monosome can be easily distinguished as a deeply staining U-shaped element applied to the nuclear membrane (Plate 4, Figs. 57, 58; Plate 7, Figs. 108, 109). During the prophase of the first maturation division the arms of the U shorten and become apposed to each other, so that the monosome appears as a longitudinally split rod (Figs. 110, 111). During the

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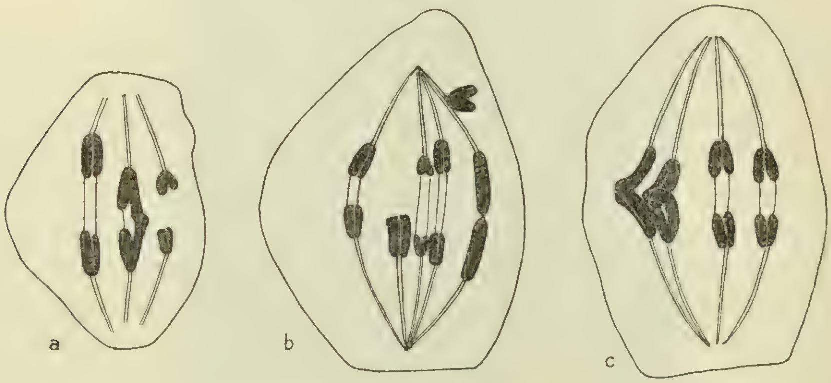
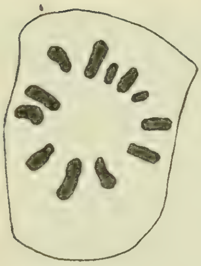
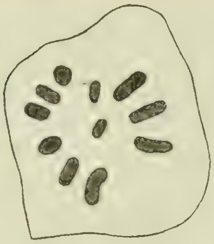
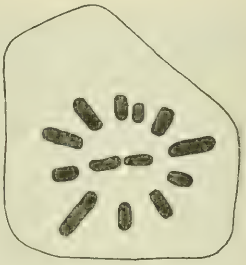




Q.



R.



S.

Figs. *Pa*, *Pb*, *Pc*.— Successive sections of spermatocyte of *Arphia tenebrosa* during the early anaphase of the first division. All the chromosomes are shown. In Fig. *Pb* are two monosomes, neither of which is dividing. × 966.

Fig. *Q*.— Polar view, metaphase of second maturation division of *Dissosteira carolina*. The monosome is present, as the total number of chromosomes (12) indicates. × 966.

Fig. *R*.— Polar view, metaphase of second maturation division of *Dissosteira carolina*. The monosome is lacking as there are only eleven chromosomes. × 966.

Fig. *S*.— Polar view, metaphase of second maturation division of *Arphia tenebrosa*. Two monosomes are present. × 966.

metaphase of the first division the monosome may lie either in the plane of the equatorial plate midway between the spindle poles or nearer one pole than the other (Plate 4, Fig. 63). Even when it lies in the equatorial plane, the monosome can be easily distinguished by its rough contour, and also by the fact that it usually lies with its axis at an angle to the axis of the spindle. In all cases the monosome is attached to spindle fibers from only one pole. The monosome does not divide during the first maturation division, but passes bodily to one of the poles (Fig. 65). In Figures *Ma–Mc* (p. 96) which are all drawn from sections of the same cell, this is conclusively shown. Here there can be no doubt that all the chromosomes except one have divided into two equal parts, which are moving toward opposite poles of the spindle. The other chromosome, the monosome, has no corresponding mate moving toward the opposite pole. As the monosome passes toward the pole the arms of the U separate somewhat, so that it has practically the same shape as the autosomes. In the succeeding telophase the monosome cannot be distinguished, but in the "semiresting" stage of the secondary spermatocyte it can again be recognized. During this stage the monosome retains its compact structure in striking contrast to the autosomes. During the second maturation division the monosome is not distinguishable, since the autosomes also have at this time somewhat rough contours. Figure *R* (p. 100) is a polar view of the equatorial plate of a secondary spermatocyte which lacks the monosome, there being only eleven chromosomes present. Figure *Q* is a similar view of a cell with twelve chromosomes, one of which is evidently the monosome. During the second division the monosome divides along with the autosomes. This is conclusively shown in Plate 5, Figures 78 and 79, which are drawn from two sections of the same cell. In this case there can be no doubt that there are twelve daughter chromosomes passing to each pole of the spindle.

The question at once arises, are we to consider the division of the monosome during the second maturation division longitudinal or transverse. Otte (:06) concludes that in Locusta, where the monosome has a somewhat similar history, the division is transverse. At first I was inclined to interpret the division in Dissosteira in the same way, but on further consideration believe such a conclusion to be unwarranted. The peculiar manner in which the monosome becomes converted into a loop during the latter part of the growth period can be easily interpreted as due to a longitudinal splitting. This interpretation is supported by the fact that, whereas the autosomes become converted into a *single* chromatin thread, the monosome becomes converted into a double thread, and at a much later period, after the autosomes have become longitudinally split. We may, then, I believe, look on the U-shaped monosome of the late growth and maturation periods as derived by a longitudinal splitting of the flattened plate of the earlier growth stages, the equivalent halves resulting from this longitudinal splitting remaining connected with each other at one end during the first maturation division, but becoming separated during the second maturation division. This interpretation is borne out by Stenobothrus, where the division is certainly longitudinal.

B. Arphia tenebrosa.

The maturation period in Arphia is practically the same as in Dissoteira.

Figure 64 (Plate **4**) shows the metaphase of the first maturation division in a cell containing two monosomes. These elements are plainly seen, one near each pole of the spindle. However, this condition is not by any means constant, since in many cases both monosomes are near the same pole. Figures *Pa–Pc* (p. 100) show three successive sections of the same cell during the early anaphase of the first division, all the chromosomes being included. These sections show conclusively that there are two monosomes, neither of which divides during this division, and each is attached to mantle fibers from only one pole. In this particular cell the monosomes are passing to opposite poles of the spindle. Figure *S* is a polar view of the metaphase of the second division in a cell which evidently contains both monosomes, since there are plainly thirteen chromosomes in the equatorial plate. Both monosomes divide longitudinally during this division, as does the single one in normal spermatocytes.

It is obvious that in the case of the individual with two monosomes the spermatids might contain eleven, twelve, or thirteen chromosomes.

C. Hippiscus tuberculatus.

The maturation period in this species differs little from the same period in Dissosteira.

D. Chortophaga viridifasciata.

The maturation period in Chortophaga agrees in all essential respects with that described for Dissosteira.

E. Melanoplus femoratus.

During the maturation period bivalent autosomes of the same types as in Dissosteira can be recognized and evidently divide in the same way.

In the case of the monosome, which at the end of the growth period is divided into two distinct components — one more or less rounded with smooth contours, the other elongated and granular — both components become homogeneous and V-shaped during the early maturation period (Plate **7**, Figs. 120–122). Finally in the late prophase of the first maturation division the components probably become joined end to end to form a longitudinally split rod. As in the preceding species, the monosome fails to divide during the first division. Figure 74 (Plate **5**) shows the metaphase of the first division. The monosome, although lying in the equatorial plane, is evidently attached at its end to mantle fibers from only one pole, while the opposite end curves away from the spindle. During the second division the monosome divides longitudinally.

F. Stenobothrus curtipennis.

1. *Autosomes.*

In Stenobothrus the tetrads are formed much as in Dissosteira, but the structure and division of the autosomes are especially well shown. At the beginning of the maturation period (stage *g*, Plate **6**, Fig. 83) the loops of the polar spireme have become freed from the nuclear membrane and the longitudinal splitting is very distinct, though never as wide as in Dissosteira. In stage *h* (Figs. 84, 85) each loop has become converted into a definitive tetrad. There are three especially large tetrads whose structure is very well shown, and these are the only autosomes I shall follow through the maturation period. The smaller autosomes are essentially like those described in Dissosteira and exhibit no new features. The three larger autosomes are, however, of especial interest, since they show the sequence of the maturation divisions in a very conclusive way. These three elements are evidently formed by the conjugation of the three pairs of larger univalent autosomes of the spermatogonia. At first they are much longer than the diameter of the nucleus, and each is plainly composed of two longitudinally split arms, which lie close together and are often more or less twisted around each other.

In stage *g* (Fig. 83) several of the free loops are clearly much longer than the others, but it is impossible to determine their number on account of their great length and the crowded condition of the nuclear elements. However, there is every reason to believe that their number is the same as that of the large tetrads which appear later, since at a little earlier stage the number of polar loops in the nucleus is undoubtedly one half that of the spermatogonial autosomes. In Stenobothrus I believe the evidence is well nigh conclusive that the large tetrads are formed by the opposite arms of the loops approaching each other, and not by an opening out of the longitudinal split. During stage *g* the longitudinal split is never very wide and later (stage *h*), when the definitive tetrads can first be distinguished, each arm has a distinct longitudinal split. In fact there is no time, until the very late prophase of the first maturation division, when the longitudinal split is not plainly discernible. Although the structure of these tetrads is not well shown at this early stage on account of their large size and the consequent bending, yet they are undoubtedly to be considered modifications of the crossed loops of Dissosteira, the differences in shape being no doubt chiefly due to their much greater size.

The autosomes rapidly shorten, thicken and become more compact, so that the structure of the three larger elements can be easily made out (Figs. 86, 87). They now show plainly their loop-like structure, the arms sometimes being nearly parallel, but more often twisted around each other; the free ends of such twisted autosomes often come together (Figs. 86, 87; Plate **7**, Figs. 183–185). In all cases, however, when strongly decolorized each can be seen to be longitudinally split, showing conclusively that the space between the arms separates univalent autosomes and therefore is not the longitudinal split. During the late prophase of the first maturation division the large bivalent autosomes become arranged on the spindle with their long axes at right angles to the spindle axis, while the spindle fibers become attached, not at the ends of the arms as in Dissosteira, but at a point some distance from the ends or the middle. This method of attachment of the spindle fibers is of especial interest, since in the spermatogonia in the case of one of the three pairs of large autosomes the spindle fibers are attached at the center of the V-shaped element, while in the case of the other two pairs they are attached at a point nearer one end than the other. Owing to their more irregular shape it is obviously impossible to determine with equal accuracy the attachment of the spindle-fibers to the larger bivalent autosomes during the first division, but after careful study I feel convinced that the same rule holds here. Figures 88, 89 are drawn from two sections of the same cell during the metaphase of the first division, all the chromosomes being shown, while Figures 90, 91 are two successive sections of another cell in the same stage and likewise show all the chromosomes. In both cells the spindle fibers are evidently attached at the middle of the arms of one of the larger autosomes, while in the case of the remaining two large autosomes the attachment is nearer one end than the other.

In the first maturation division, as we should expect, there are, in addition to the three larger autosomes, five smaller autosomes in the equatorial plate, which show approximately the same size relations as the autosome pairs of the spermatogonia (Fig. *NN*, p. 96).

As a consequence of the manner of attachment of the spindle fibers described above, the first division must result in the separation of the arms of the loop-shaped, bivalent autosomes. Although apparently longitudinal in the case of the three larger autosomes, it is nevertheless a true reducing division, since, as we have seen, each arm represents a univalent autosome. As the univalent components separate they assume the usual V-shape, but in the case of the three larger autosomes the space between the arms of the Vs does not represent the longitudinal split as in the smaller autosomes. As the V-shaped univalent components of the larger autosomes separate (Fig. 93) each is clearly double owing to the reappearance of the longitudinal split. Occasionally one of the large autosomes may become so placed on the spindle that the arms instead of lying on the spindle throughout their entire length as usual, project away at an angle (Plate **7**, Fig. 187). In such cases it is the free ends of the arms which separate first, giving rise to characteristic E-shaped forms (Fig. 188; Plate **6**, Fig. 91). Figure 94 shows the end of the anaphase of the first division. As in the preceding species, there is in the secondary spermatocytes a partial resting stage (Fig. 95), which lasts but a short time.

As the chromosomes become drawn into the equatorial plate of the second division figure (Fig. 96) the three larger autosomes form double Vs and, as in the first division, the attachment of the spindle fibers is near the middle of the V-shaped elements. Figures *T* and *U* show that, as in the first division, in two of the large autosomes the spindle fibers are attached nearer one end than the other, while in the third large autosome this attachment is at a point directly in the center. Thus we have in each of these autosomes a characteristic method of attachment to the spindle fibers, which persists through the spermatogonial and maturation divisions. During the second division the autosomes divide along the plane of the longitudinal split which first appeared in the polar spireme of the primary spermatocyte. The second maturation division is therefore an equational division. Figure 97 shows the anaphase and Figure 98 the telophase of the second maturation division.

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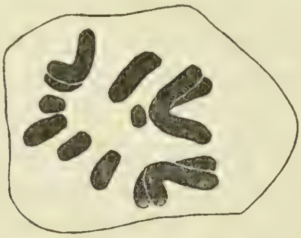
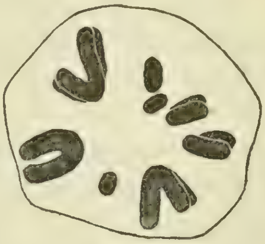
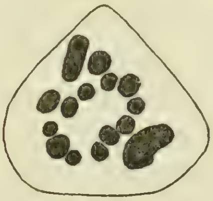
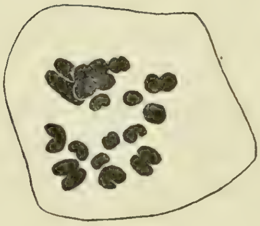


U.

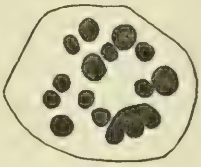






V.

W.



X.

Fig. *T*.— Polar view, metaphase of second maturation division of *Stenobothrus curtipennis*. Nine chromosomes. The monosome is present. × 1450.

Fig. *U*.— Polar view, metaphase of second maturation division of *Stenebothrus curtipennis*. Eight chromosomes. The monosome is absent. × 1450.

Fig. *V*.— Polar view of daughter chromosomes, anaphase of first maturation division in *Steiroxys trilineata*. × 1450.

Fig. *W*.— Polar view, metaphase of second maturation division in *Steiroxys trilineata*. Fifteen chromosomes. The monosome is present. × 1450.

Fig. *X*.— Polar view, metaphase of second maturation division in *Steiroxys trilineata*. Fourteen chromosomes. The monosome is absent. × 1450.

2. *Monosome.*

It still remains to follow the monosome of Stenobothrus through the maturation period. At the end of the growth period this element is plainly composed of two distinct parts (Plate **3**, Fig. 52), one of which is rounded, has smooth contours, and is vacuolated, while the other is elongated, has a ragged outline, and is granular. In the following stage (*g*) the granular part shortens and becomes more compact, meanwhile being bent into a V-shape (Plate **7**, Figs. 140–141).

The other part of the monosome assumes a similar shape, the change being apparently brought about by the development, near the center, of a thin area which finally breaks through forming a ring. A further extension of this thinning process through one side of the ring results in its interruption and the formation of a V-shaped body. This leads to the next stage (Figs. 143–145) in which both parts of the monosome are distinctly V-shaped, although they can still be distinguished from each other since one has a rough contour while the other is perfectly smooth. The two components of the monosome usually lie close together, but may be separated a short distance, as in Figure 142, although in such cases they are always connected by linin fibers. The components, as indicated in the figures, may be differently oriented in relation to each other, in some cases lying side by side, in others end to end. In the latter case the monosome often strikingly resembles a tetrad (Fig. 145). The further changes consist in a shortening and thickening of the two components, which usually fuse end to end, the space between the arms of the Vs becoming gradually obliterated (Figs. 146–148). Occasionally the components may lie side by side (Figs. 149, 150), but apparently in such cases they are later connected at only one end. Usually during the metaphase of the first maturation division (Plate **6**, Figs. 88, 91, 92) the monosome has the form of a straight rod,— the two components being indistinguishable,— but rarely it may be more or less curved. Probably such curved rods are derived from forms like those shown in Figures 149 and 150.

During the metaphase of the first division the monosome usually lies nearer one spindle pole than the other (Plate **6**, Figs. 88, 91, 92), and, as in the preceding species, is easily distinguished by its more ragged outline. During the anaphase (Fig. 93) the monosome does not divide, but passes bodily to one pole. At this stage it shows a distinct longitudinal split. In the secondary spermatocyte (Fig. 95) the monosome is easily recognizable by its more compact structure. Throughout this stage it shows a distinct longitudinal split. In the second maturation division the monosome can be easily distinguished by its relative size. Figure *U* (p. 106) is a polar view of the equatorial plate in a cell which lacks the monosome, while Figure *T* is a similar view of a cell containing this element. During the second maturation division the monosome divides longitudinally and consequently one half of each of the two components passes into each daughter cell.

In Stenobothrus, as in the preceding species, there is a distinct dimorphism of the spermatids, one half containing the monosome, while the other half lacks this element. The monosome (Plate **5**, Fig. 82) is easily distinguishable in the spermatid for some time, as its disintegration takes place much later than that of the autosomes.

G. Steiroxys trilineata.

1. *Autosome.*

At the close of the growth period the loops of the polar spireme become freed from the nuclear membrane, as in the Acrididae. Moreover, the chromatin and linin become much concentrated so that in stage *g* (Plate **5**, Fig. 70) the diameter of the threads is much less than during the preceding stage. At the same time, as a result of the aggregation of the chromatin, the longitudinal split reappears. Somewhat later the loops have become converted into tetrads, which stain deeply and have such a ragged outline that it is almost impossible to determine their structure; but they have apparently (Figs. 71, 72) forms similar to those of the Acrididae. There is always one tetrad which is much larger than the others and is undoubtedly formed from the two large autosomes of the spermatogonia. The form of this tetrad is well shown in Figure 71. It is apparently formed by the arms of the loops becoming twisted around each other, and, as in the Acrididae, each of these arms no doubt represents a univalent autosome. During the late prophase of the first maturation division this autosome becomes disposed on the spindle with its long axis at right angles to the axis of the spindle (Fig. 73). In addition to the large element there are thirteen smaller autosomes (Plate **8**, Fig. 189), which have approximately the same size relations as the autosome pairs of the spermatogonia. In the case of the large autosome the following division is apparently longitudinal, but, judging from analogy with the Acrididae, results in the separation of its univalent components. As the two components separate (Fig. 190) each is split longitudinally and is evidently composed of two curved rods lying side by side. In the case of the smaller autosomes I have found it impossible to determine their orientation on the spindle with any accuracy, but, as in the large autosome, the first division is probably reducing. As the small autosomes separate (Fig. 190), their dyad structure is usually apparent, although the longitudinal split is often difficult to distinguish and can best be seen in polar views of the anaphase (Fig. *V*, p. 106). Figure 191 shows the beginning of the telophase, the autosomes being collected in a mass near the poles of the spindle, while Figure 192 is a late telophase. During the "semiresting stage" of the secondary spermatocytes the autosomes become broken down to a greater extent than in the Acrididae, and the chromatin is scattered through the nucleus in irregular masses (Fig. 193). I have, however, been unable to distinguish any nuclear membrane at this stage, although the nucleus is well defined. Figure 194 is a prophase of the second maturation division, the longitudinal split in the large autosome being clearly shown. Figure 195 shows the metaphase, and Figures 196 and 197 different stages of the anaphase of the second division. In the large autosomes this division is plainly longitudinal and equational, since it is, in all probability, along the plane of the longitudinal split. Here, again, the plane of division cannot be determined in the remaining autosomes, but is probably longitudinal. In Figures 198 and 199 is shown the beginning of the telophase of the second division.

Thus, in the case of the large bivalent autosome both divisions are longitudinal, although the first is really a reducing division. In the other autosomes the first division is probably transverse, the second longitudinal. I have been unable to find the slightest evidence that both maturation divisions are transverse, as is held by Otte (:06) to be the case in Locusta.

2. *Monosome.*

The monosome in Steiroxys, unlike that in most of the Acrididae, can be easily followed throughout both maturation divisions. At the beginning of the maturation period the monosome, which in the preceding stage formed a flattened vacuolated plate, becomes more compact and is soon converted into a U-shaped element (Plate **7**, Figs. 155–158) by the development of a longitudinal split. It is interesting to note that the monosome has at this time practically the same form as during the early growth period, except that it is now much shortened and thickened. The arms of the U become closely apposed and during the metaphase of the first maturation division (Plate **5**, Fig. 73) the monosome is seen as a longitudinally split rod lying nearer one spindle pole than the other. It usually lies in the cytoplasm at a short distance from the spindle, but in favorable cases can be plainly seen to be connected by mantle fibers with the nearest pole. As in the Acrididae, the monosome (Plate **8**, Fig. 190) does not divide during the first division. In the majority of cases it passes to one pole of the spindle with the autosomes, but occasionally it lags behind (Fig. 191). In such cases it never enters the nucleus of the secondary spermatocyte, but lies outside in the cytoplasm, where it forms a conspicuous U-shaped element. Figure 193 represents two adjoining secondary spermatocytes during the "semiresting stage"; in one of these the monosome lies in the cytoplasm, while in the other it is located within the nucleus. Similar conditions have been described by Baumgartner (:04) and Gutherz (:07) in Gryllus, and by Otte (:06) in Locusta.

During the second maturation division the monosome can always be easily recognized by its relative size. Figure *W* (p. 106) shows an equatorial plate of this division with, and Figure *T* one without, the monosome. During the second division the monosome divides longitudinally, one arm of the U going to each pole (Fig. 197), but it always lags behind the autosomes. During the early telophase the monosome can always be recognized projecting out from the mass of chromosomes which are collected at each pole of the spindle (Fig. 199). Figure 198 shows the same stage where no monosome is present.

In the spermatids the monosome retains its compact structure until a much later stage than in the Acrididae, being recognizable up to a late stage in the metamorphosis. However, it is, of course, present in only one-half of the spermatids.

5. The Metamorphosis of the Spermatids.

A. Dissosteira carolina.

The metamorphosis of the spermatids appears to be practically the same in all the Acrididae, so I have thought it necessary to describe the process in only one species. During the telophase of the second maturation division (Plate **5**, Fig. 80) a nucleus is formed in each cell, as usual. The autosomes rapidly disintegrate and become scattered through the nucleus as fine granules suspended in linin meshwork (Fig. 81). The monosome, however, does not break down until later, but forms a conspicuous element in one-half of the spermatids, where it appears as an elongated deeply staining body immediately within the nuclear membrane (Fig. 81). For some time the spermatids are connected by the interzonal filaments of the second maturation division, which are at first very conspicuous but gradually disappear. Meanwhile, a deeply staining mass has appeared by the side of the nucleus at the end of the interzonal filaments. This body, which is evidently the Nebenkern, is at first irregular in shape, very finely granular, and stains deeply with Bordeaux. Apparently it is derived chiefly from the mitochondrion (which it closely resembles), although the interzonal filaments may also be concerned in its formation.

However, practically the only evidence for this is the fact that as the Nebenkern develops the interzonal filaments gradually disappear. Certainly these filaments do not become converted directly into the Nebenkern, but if they take part in its formation must first disintegrate. Figure 214 (Plate 9) represents a spermatid in a somewhat later stage after the Nebenkern is fully formed. In this case the interzonal body is still faintly distinguishable, although no trace is left of its fibrillar structure. Usually the interzonal body is not recognizable at so late a stage. Figure 213 shows a spermatid in which the Nebenbern is still very irregular. The nucleus has lost its staining power to a considerable extent, and its appearance is quite different from that at a little earlier stage, but is characteristic of this and later stages. The entire nucleus has a finely granular structure without any well defined network or differentiation of chromatin and linin. The fine granules stain lightly with hematoxylin, so that the nucleus as a whole appears grayish, with here and there a few large granules staining deeply with hematoxylin. The monosome is still distinguishable as a compact, deeply staining body. For some time the further changes in the nucleus consist chiefly in the gradual disintegration of the larger deeply staining chromatin granules, including the monosome, until finally all the chromatin becomes converted into fine granules which have lost to a very large extent their affinity for hematoxylin (Figs. 215–219). In Fig. 214 the monosome has begun to disintegrate, while the Nebenkern forms a rounded, homogeneous, deeply staining body by the side of the nucleus. It already shows indications of dividing into two equal parts by constriction. The cytoplasm on the side containing the Nebenkern is just beginning to grow out to form the tail of the spermatozoön. In Figure 215 the Nebenkern has divided into two equal parts. During this and slightly earlier stages it often shows near the periphery a very narrow lighter area outside of which is a thin envelope of more deeply staining material. Probably this appearance is due to faulty fixation, since it is more marked in poorly fixed material and since in some cases, especially near the outer walls of the follicle, where the cells are most easily acted on by the fixing agent, the Nebenkern appears perfectly homogeneous. It is an interesting fact, which has also been noted by other writers, that the spermatids seem to be the most difficult elements in the testis to fix properly.

By the time the Nebenkern has divided, the entire cell has become considerably elongated. Applied to the exterior of the nuclear membrane can be seen a small deeply staining body which is evidently the "centrosome" of writers on spermatid metamorphosis. I have been unable to trace any connection between this body and the centrosome of the last maturation division since, as in previous divisions, the centrosome disappears during the anaphase. During the early stages of the metamorphosis granules staining with hematoxylin are frequently seen in the cytoplasm, and in some cases may be applied to the nuclear membrane. There may be several of these granules in a single spermatid, and it is, of course, impossible to determine whether any of them have any connection with the centrosome. Extending out from the centrosome into the elongated portion of the cell is a fine fibril, the axial filament. At this stage the axial filament is well defined for a short distance from the centrosome and then gradually becomes thinner until it disappears altogether. The spermatid continues to elongate (Fig. 216) while the centrosome and axial filament increase in size. The Nebenkern becomes much elongated and travels out along the axial filament, part of it being continually left behind to form a distinct envelope about the filament. Figure 218 is a later stage, in which the entire Nebenkern has been converted into an envelope surrounding the axial filament. The nucleus lies at one end of the greatly elongated spermatid and is surrounded by a very thin layer of cytoplasm.

From now on marked changes take place in both cytoplasm and nucleus. The latter becomes smaller and conical, and stains a nearly uniform gray with here and there minute deeply staining granules (Fig. 219). Figures 220–222 represent successively later stages in the elongation of the nucleus and centrosome. At this time, especially when imperfectly fixed, the nucleus usually shows a very finely fibrillar structure. There is practically no cytoplasm surrounding the nucleus, although it is still plainly enclosed by the cell membrane. Figure 223 shows the anterior end of a nearly mature spermatozoön. The nucleus, which again stains deeply with hematoxylin, has become greatly elongated to form the head of the spermatozoön and at the anterior end tapers to a fine point; but there is nothing which can be considered an acrosome. The nucleus is still surrounded by the cell wall, but there is apparently no cytoplasm in this region and the cell wall is probably lost a little later. The centrosome is much elongated and forms the so-called middle piece, while the tail is composed of a central deeply staining fiber, formed from the axial filament, and an envelope derived from the Nebenkern, the two at this stage being indistinguishable. Surrounding the central fiber and forming the greater part of the tail is a lighter envelope derived from the cytoplasm of the spermatid. The further changes consist chiefly in a still greater elongation of the head until it becomes several times the length shown in Figure 223 and correspondingly smaller in diameter.

Spermatids which are much larger than normal ones and contain two or more centrosomes are occasionally met with. Figure 217 shows one of these abnormal spermatids with four centrosomes, each of which has an axial filament connected with it. The number of such abnormal spermatids varies in different individuals, although they are never very common. Similar abnormal spermatids have been described by Paulmier ('99) and others.

I have not found evidence of the degeneration of spermatids or spermatozoa, except in rare cases.

Inasmuch as only one-half of the spermatids contain the monosome, and as there is no evidence that the monosome is extruded from the nucleus during the metamorphosis or that any considerable number of the spermatids degenerate, it follows that the mature spermatozoa are dimorphic, even though there is no recognizable difference between them,— half of them containing the substance of the monosome, the other half being without that substance.

B. Steiroxys trilineata.

In the resting spermatids (Plate 8, Figs. 200–203) the autosomes have become broken down into granules distributed through the nucleus, but the monosome retains its compact form and shows the staining reactions characteristic of dividing chromosomes. It forms a lenticular body closely applied to the inner surface of the nuclear membrane and, on account of its large size and staining reactions, is a very conspicuous element of the spermatids. Figures 200 and 202 show resting spermatids which contain the monosome, while Figures 201 and 203 show others in the same stage in which this element is lacking. Lying near the nucleus, on the side covered with the greatest amount of cytoplasm, is a rounded homogeneous body staining deeply with Bordeaux; for convenience I shall call this the Nebenkern, without implying anything in regard to its homologies. Often a less deeply staining body can be distinguished closely applied to the nuclear membrane near the Nebenkern (Fig. 203). This body is usually difficult to distinguish from the surrounding cytoplasm, but is probably of universal occurrence. During the telophase of the second maturation division the cytoplasmic structures are very indistinct, so that I have been unable to determine the origin of the Nebenkern and the neighboring body. At a little later stage (Fig. 204) the spermatid has become much elongated and an axial filament is present. I have been unable to find, at this time, any trace of a centrosome, but the axial filament appears to be attached directly to the nuclear membrane at a point nearly, or quite, 90° from the Nebenkern. The small, less deeply staining body which was formerly applied to the nuclear membrane near the Nebenkern can no longer be distinguished. Probably it has become converted into the homogeneous envelope which surrounds the axial filament.

The Nebenkern now presents a very striking appearance, since one side stains intensely with hematoxylin. However, the staining qualities of the Nebenkern during this and later stages appears to be dependent on the quality of the fixation. While one side of the Nebenkern stains deeply in spermatids exhibiting the best fixation,— e. g. those lying near the follicular wall,— in the case of spermatids less perfectly fixed the stain is less intense, or may be absent altogether. Also, in material fixed in Worcester's formol-sublimate-acetic fluid the Nebenkern stains lightly or not at all.

The chromatin continues to disintegrate into finer granules, although the monosome still retains its characteristic structure. Somewhat later (Fig. 205) the finely granular chromatin is collected around the periphery of the nucleus, while at the center there is a lighter region apparently free from chromatin. However, the peripheral distribution of the chromatin persists only a short time, for a little later it is seen to occupy the anterior end of the nucleus, while at the posterior end there is a lighter region (Fig. 206). The chromatin, including the monosome, has now become converted into very fine granules, which stain with hematoxylin less deeply than formerly, so that, as in Dissosteira, the nucleus as a whole stains grayish. The nucleus now rotates through an angle of about 90° and at the same time two minute deeply staining centrosomes appear at the proximal end of the axial filament, where it is attached to the nucleus. I have not been able to determine the origin of these centrosomes nor whether, indeed, they lie within or without the nuclear membrane, to which they are closely applied. Meanwhile, the Nebenkern begins to migrate toward the anterior end of the nucleus. The deeply staining portion has now spread over one side of this body, so that about one-half of the surface of the Nebenkern is seen to be enveloped by a deeply staining cap. In Figure 207 the deeply staining cap has extended over the entire surface of the Nebenkern, which has now become applied to the anterior end of the nucleus. The further changes in the spermatids are chiefly due to an elongation and flattening of the nucleus, while at the same time the Nebenkern extends back for some distance along the sides of the nucleus and goes through a complicated metamorphosis. In Figure 208 the Nebenkern is seen to form a deeply staining cap closely applied to the anterior end of the somewhat flattened nucleus, the sides of which it also envelops for a greater or less distance. Figure 209 shows little change except that the nucleus is more elongated. Figure 210 represents a later stage, in which the nucleus has become still more elongated and flattened so that it may be compared to a paddle, the tail corresponding to the handle. Each centrosome has divided, so that four deeply staining granules can be seen at the end of the axial filament. However, the most striking change has taken place in the anterior cap derived from the Nebenkern, which becomes converted into the acrosome. It no longer stains uniformly, but appears as an elongate deeply staining body on each side of the anterior end of the nucleus. The median portion of the cap stains only slightly or not at all with hematoxylin, except along its anterior margin, where there are two rows of minute deeply staining granules, one row lying slightly posterior to the other. Figures 211 and 212 represent a still later stage when the nucleus takes a nearly uniform gray stain, but appears black when viewed from the side, owing to its greater thickness in that direction. Both nucleus and acrosome appear considerably larger as a result of their having become much more flattened. The granules at the anterior end of the acrosome are much more conspicuous than formerly and can be seen to be arranged in the form of an ellipse the anterior and posterior sides of which lie at slightly different levels. Figures 211 and 212 give a much better idea of the structure than can be conveyed by an extended description.

The cell wall can still be distinguished surrounding the nucleus but, as in Dissosteira, I have been unable to find any cytoplasm in this region, although it forms a distinct envelope around the axial filament.

This is the latest stage in the metamorphosis which I have been able to find in my preparations; but the metamorphosis is evidently nearly completed.

As in Dissosteira, there are undoubtedly two varieties of spermatozoa with respect to their chromatin contents; one half of them containing the monosome while the other half lack this element; the two types show, however, no external differences.

IV. Discussion.

1. The Apical Cell.

The apical cell is a large element of characteristic appearance. It occurs at the distal ends of the follicles quite generally in the testes of insects, and also, in some cases at least, in their ovaries. This cell was formerly known as Verson's cell, after its supposed discoverer; but, as Cholodkovsky (:05) points out, it was in reality first described by Spichardt ('86). There would therefore seem to be no good reason for retaining the name, and I have preferred to use the more appropriate term of *apical cell* given it by Grünberg (:03).

The apical cell was first found and has been chiefly studied in the Lepidoptera, where it is the most conspicuous element in the entire testis. The following authors have described the apical cell in this group of insects: — Spichardt ('86), Verson ('89, '91, '94), Cholodkovsky ('94), Toyama ('94), La Valette St. George ('97), Tichomirow ('98), Grünberg (:03), and Munson (:06). Toyama, La Valette St. George and Grünberg also found a similar cell in the ovary.

In Lepidoptera the apical cell, besides being much larger than the other elements of the testis, has an irregular outline. Near the center of the cell is a large nucleus, while distributed through the cytoplasm, especially in the vicinity of the nucleus, are large numbers of deeply staining granules. The cell is closely surrounded on all sides by several concentric layers of spermatogonia.

Cholodkovsky ('92, '94, :05) has found the apical cell in the testes of several Diptera, a hemipteron (Syromastes) and a neuropteron (Phryganea), while Holmgren (:01) has found it in a coleopteron (Staphylinus).

It seems not to have been previously reported for the Orthoptera, although it was evidently seen by Sutton (:00), who supposed it to be a degenerating spermatogonium which had become inclosed by the fusion of two neighboring cysts.

This cell seems to be of universal occurrence in the Acrididae and Locustidae, as I have found it in a large number of species belonging to both families. In no case have I failed to find the apical cell in the usual position, although in some species it is much more conspicuous than in others. I have also found a similar cell in the testes of a cricket (Gryllidae) and a cockroach (Blattidae), so it probably occurs throughout the Orthoptera.

The apical cell seems not to occur in other groups of animals than insects, although Lerat (:05) has found peculiar cells in the ovary and testis of Cyclops which seem to agree with it in some respects.

Regarding the function of this cell there are in general two views: one, that it is the progenitor of the germ cells; the other, that it has nothing to do with the origin of the germ cells, but functions simply as a supporting or nurse cell. Verson ('89, '91, '94) was one of the chief exponents of the former view, holding that the other elements of the testis are derived from the apical cell by amitotic division. Cholodkovsky formerly ('94) held the same view as to the origin of the testicular elements, but believed the divisions to be mitotic. His later views are mentioned further on. More recently this view of the function of the apical cell has given way to the belief that it is a supporting element; however, Verson's interpretation has been revived by Munson (:06). This author believes that the apical cell (which he calls the "grandmother stem cell") gives rise in some way to his "mother branch cells," which immediately surround, and are in close connection with the apical cell. These "mother branch cells" undergo repeated mitotic divisions, the peripheral (distal) one of the daughter cells of each division being budded off to form a "primary spermatogone," which is the progenitor of all the spermatogonia of a given cyst, while the other, or proximal one, remains connected with the apical cell to give rise later to successive primary spermatogonia. The cell which is separated off (primary spermatogone) is accompanied by one or more small nuclei, which later develop into cyst cells. These nuclei Munson believes to be derived from very minute granules which occur in the peripheral cytoplasm of the apical cell. They are apparently the same granules which have been interpreted by other investigators as metabolic products. Just why Munson believes the spermatogonia to be derived from the apical cell is not apparent, since he admits that he has never seen this cell undergoing division, and in all cases observed by him it differed greatly in appearance from the spermatogonia.

Turning now to the other view,— that the apical cell is a supporting or nurse cell,— we find that this interpretation has been accepted in some form or other by most authors. This view was held by Ziegler und Vom Rath ('91), Toyama ('94), Erlanger ('96), La Valette St. George ('97), Tichomirow ('98), Holmgren (:01), Grünberg (:03), and recently Cholodkovsky (:05). Holmgren (:01) has traced deeply staining granules from the nucleus into the surrounding cytoplasm. He also found granules in the cytoplasm of the surrounding spermatogonia staining like those of the apical cell. Grünberg (:03) has given a detailed account of the apical cell in a number of Lepidoptera and has come to the conclusion that it has two, more or less distinct, functions. He agrees with La Valette St. George, in opposition to Toyama, that it is a germ cell, but one which has become modified for an entirely different function. In the embryo and young larva the apical cell is closely applied to the testicular wall at the distal end of the follicle. Later the cell enlarges and between it and the surrounding spermatogonia can be seen a lighter area filled with cytoplasm containing numbers of deeply staining granules. He believes this area to be formed by the disintegration of primary spermatogonia which were directly in contact with the apical cell, and that the remains of these cells, after being elaborated by the apical cell, serve as nutriment for the remaining spermatogonia. Somewhat later the apical cell moves into the lumen of the follicle, but for a time remains attached to an ingrowth of the testicular wall. This connection with the outer wall of the testis is probably retained to enable the cell to procure nutritive material, which it elaborates for the use of the spermatogonia. This view is supported by the fact that the cytoplasmic granules often show a radiate arrangement, as though streaming out from the nucleus to the surrounding spermatogonia. Grünberg (:03, p. 378) concludes: "Ihre Thätigkeit als solche kann eine doppelte sein: durch Aufnahme von Material und Verarbeitung desselben übt sie eine *assimilirende* Thätigkeit aus; ausserdem kann sie durch selbständige Produktion von Nährsubstanz die Bedeutung einer *secernirenden Nährzelle* gewinnen." Recently Cholodkovsky (:05) has changed his former view and accepts Grünberg's conclusions.

In the Orthoptera the appearance of the apical cell suggests that it has a similar function, although the evidence is far from conclusive. It is possible that this cell is concerned in the formation of the mitochondrion, since this substance in the primary spermatogonia closely resembles the finely granular material surrounding the nucleus of the apical cell. In this connection it is interesting to note that the primary spermatogonia, although dividing rapidly, are always of approximately the same size, while the secondary spermatogonia rapidly decrease in size in the later generations.

2. The Spermatogonial Autosomes.

It has long been known that there is often considerable variation in the size of the different chromosomes in the same species, but it was formerly supposed that such differences are largely accidental and not of fundamental importance. It is only within recent years that the significance of the variation in the size of the chromosomes of the same species has been appreciated. Montgomery (:01) first showed that in several Hemiptera some of the autosomes in the spermatogonia are distinguishable by their size, and that there are always two of each size. He also showed that during the maturation divisions the autosomes so divide that each spermatid contains one member of each pair. Since probably a similar process occurs in oögenesis, Montgomery concluded that one of each pair of autosomes is derived from either parent. In the following year Sutton (:02) elaborated this idea at considerable length. Sutton found that in the spermatogonia of Brachystola magna there are twenty-two autosomes, which can be arranged in pairs according to their size. He agreed with Montgomery that during maturation the members of each pair become separated, so that in the gametes there is only one autosome of each size. The fusion of the male and female gametes during fertilization results in a restoration of the original paired condition. Recently a number of investigators have found similar conditions in the germ cells of animals belonging to widely separated groups, but the autosome pairs appear to be most marked in the insects, where they are often shown with almost diagrammatic clearness. Baumgartner (:04) found that in Gryllus the autosomes can be arranged in graded pairs, although in some cases the difference between the pairs is very slight. A similar result was reached by Montgomery (:05) in Syrbula. In this species there are twenty chromosomes in the spermatogonia, which are evidently paired, and he was able to distinguish the three largest and three smallest pairs, but the remaining eight chromosomes are so nearly of the same size that the individual pairs could not be distinguished. Montgomery believed one of these medium sized pairs to be allosomes. Stevens (:05) has shown that in two species of Aphis the ten autosomes can be readily grouped in five pairs and she later (:06) obtained similar results in both sexes of a number of additional species. In these insects, owing to the small number of autosomes and their great variation in size, the paired relation is shown with exceptional clearness. Wilson (:05^a) has found a similar condition in the spermatogonia of several Hemiptera and he (:06) has described in detail the autosome pairs in a number of additional forms. He also found a similar series of paired autosomes in the oögonia, and even in the follicle cells of the ovary; likewise in the investing cells of the testis there is the same or a multiple series of autosomes. These results have been confirmed in the case of the spermatogonia by Montgomery (:06), who has investigated a large number of Hemiptera from this standpoint. Montgomery also noted that the members of each pair are usually closely associated during mitosis. In this paper he goes further than any of the preceding authors and claims that while the autosomes are evidently paired, yet the components of each pair are never of exactly the same size. This he believes to be due to the fact that the chromosomes derived from the female parent are always slightly larger then those from the male parent. That it is possible to distinguish such slight differences in size, appears to me in the highest degree improbable. Even though exactly alike, two chromosomes would rarely appear of precisely the same size, owing to slight variations in fixation, or staining, or to different degrees of foreshortening, etc. It is indeed probable that the components of a pair often differ slightly in form, volume, etc., yet there seems to be no good reason for believing that these slight differences persist from generation to generation. During the resting stage each chromosome becomes broken up into fine granules, which double in volume before the next division, and it would seem to be asking too much of the theory of the individuality of the chromosomes to expect each chromosome to be reconstituted with *exactly* the same form and volume as in the preceding division. Moreover, Montgomery's reason for considering that the smaller chromosomes are always derived from the male parent does not seem to be well founded. It is based entirely on analogy with the diplosomes, where it is probable that in the case of the male germ cells the smaller diplosome is derived from the male parent. Montgomery apparently forgets that in the female the diplosomes are of equal size although one is of paternal, the other of maternal origin.

Otte (:06) has found that in the spermatogonia of Locusta the autosomes are paired, and Zweiger (:06) has shown that the same is probably true of Forficula.

In other groups similar results have been obtained by a number of recent writers, although in many cases the individual pairs are not as marked as in the insects. Among the arachnids the autosomes of Lycosa have been shown to be paired by Montgomery (:05) and those of Epeira by Berry (:06). In the worms, Montgomery has found that in the first cleavage spindle of Ascaris megalocephala two of the four autosomes are somewhat larger than the others. A. und K. E. Schreiner (:06) found that in Tomopteris there are eighteen autosomes in the spermatogonia, two being considerably shorter than the others, which are also probably paired, although the difference in size is slight. In the gastropods we have the observations of Bonnevie (:05, :06), who found in the oögonia thirty-four autosomes, which can be divided according to their size into three groups, each group containing an equal number. She also observed slight variations in the size of the elements within each group. Turning to the vertebrates, we find that similar results have been arrived at by Montgomery (:04) and by A. und K. E. Schreiner (:04, :05, :07). According to Montgomery the spermatogonial autosomes of several salamanders can be grouped in pairs of like volume and form, the components of each pair usually lying close together in the spindle. Similarly the Schreiners have shown that in the spermatogonia of Myxine and Spinax certain autosomes can be recognized by their size, and that in such cases there are always two of equal volume. They also confirm Montgomery's observations that the components of each pair usually lie close together.

My own results in the Orthoptera are in perfect accord with those of the other observers cited.

In the light of the results detailed above there would seem to be little room for doubting that in the germ cells of all animals which reproduce sexually, there is a double series of autosomes, one being of paternal the other of maternal origin; but such a series is recognizable only where there are considerable differences in the volumes of the elements. The further fact, first noted by Montgomery (:04), that the components of each pair are usually closely associated, is significant as indicating a physiological relationship between the two. Montgomery has sought to explain this association on the supposition that during the prophases the components of each pair lie close together in a continuous linin spireme. This is, of course, based on his assumption (:00) that the chromatin and linin form a single element of which the chromosomes are simply subdivisions. It is not my purpose to point out here the many, and as I believe fatal objections to this view, but to consider only its bearing on the association of homologous chromosomes. If there is any such fixed organization of the nucleus as Montgomery imagines, it would seem that the elements of each pair ought always to lie close together, but this is certainly not the case. While it is undoubtedly true that such an association is the rule, yet exceptions are common and the members may be widely separated on the spindle. This is especially well shown in Steiroxys where the elements of the largest pair can always be recognized at a glance. In this case the large autosomes usually lie close together, but may be as far apart as the entire diameter of the spindle (cf. Plate **2**, Fig. 18, and Fig. *L*, p. 77). I believe the facts can be better explained on the assumption that there is a marked attraction between the components of each pair, which results under ordinary circumstances in their lying close together, but would not prevent their being temporarily separated by various factors, such as the crowding of neighboring chromosomes, and the like.

McClung (:**05**) has held that there may be a still closer association of the chromosomes in the spermatogonia. He asserts that in several Orthoptera a "precocious conjugation" of certain autosomes may occur at this early stage. The rod-shaped autosomes become joined end to end forming a U-shaped element, to the center of which the mantle fibers become attached. I have no material from any of the species in which McClung found such precocious conjugation, with the exception of Chortophaga viridifasciata, but in this species I have been unable to find any evidence of conjugation in the spermatogonia. In fact, in my preparations of Chortophaga the chromosomes are, if anything, more widely separated than in most species. However, I have been able to find very few cells which afford a good view of the equatorial plate and these are all among the earlier generations. It is probable that in the later generations, owing to the decrease in the size of the cells, the chromosomes become more closely crowded together. It is possible that in some cases McClung has mistaken a univalent for a bivalent autosome. Apparently one of his chief reasons for considering certain autosomes in the spermatogonia to be bivalent is the fact that they are U-shaped — having the mantle fibers attached at the apex, and exhibit the so-called heterotypical form of mitosis. In Stenobothrus six of the spermatogonial autosomes show these characteristics, and yet it is very certain that they are all univalent.

3. Synapsis.

It is not my intention to attempt a complete review of the already enormous literature on this stage in the development of the germ cells, but only to consider the more important results of some of the more recent investigations. The term synapsis was first applied by Moore ('**95**) to a stage in the early growth period when the chromatin is massed at one side of the nucleus, during which, as he believed, the reduction in the number of chromosomes takes place. Inasmuch as it has since been shown that in many cases no such contraction of the chromatin occurs, the term has been used in a wider sense to apply to the entire series of phenomena which are concerned in the conjugation of the chromosomes, and especially to the stage at which the reduced number of chromosomes is first apparent. Moore himself (Farmer and Moore :05) has used the term in this somewhat modified sense, where it is equivalent to his "contraction figure," and has defined it as follows:—"Synapsis represents that series of events which are concerned in causing the temporary union in pairs of premaiotic chromosomes, previously to their transverse separation and distribution, in entirety between two daughter nuclei." Used in this sense the term will apply, in the case of the male germ cells at least, to the greater part of the growth period, and especially to the stage of the polar spireme. In fact, an examination of the literature leads to the conclusion that the occurrence of the polar loops is the most characteristic phenomenon of the synaptic period, and while in the present stage of our knowledge it does not appear to be of universal occurrence, yet this arrangement of the spireme threads is so common and occurs in such widely separated groups as to indicate that it is of fundamental importance. Indeed, I suspect it will be found to be the most characteristic stage of the synaptic period. Montgomery (:00) seems to have been the first to call attention to the fact that in the growth period of the germ cells in many animals the spireme is in the form of loops with their open ends directed toward the distal pole, where they are more or less closely attached to the nuclear membrane. Such an arrangement of the spireme during synapsis has since been described in a large number of forms. Among others in mammals by von Winiwarter (:00, :02); in amphibians by Kingsbury ('99, :02), Eisen (:00), Montgomery (:03, :04), Janssens (:05), Moore and Embleton (:06), and A. und K. E. Schreiner (:07); in fishes by Moore ('95), A. und K. E. Schreiner (:04, :05, :07), Maréchal (:04, :05), and by Farmer and Moore (:05); in insects by de Sinéty (:01), Baumgartner (:04), Montgomery (:05), Farmer and Moore (:05), Stevens (:05^a, :06^a), Nowlin (:06) and Otte (:06); in arachnids by Montgomery (:05) and Wallace (:05); in Peripatus by Montgomery (:00); in copepods by Lerat (:05); in gastropods by Meves (:02) and Bonnevie (:05, :06), and in annelids by A. und K. E. Schreiner (:06, :06^a). In all cases where the number of loops has been determined there is always, as in the Orthoptera, at least during the latter part of the polar stage, one-half as many loops as somatic autosomes. The above makes no pretence to being a complete list, but is given to show the wide occurrence of the polar loops during synapsis. I have no doubt that a reëxamination will show that a similar arrangement of the spireme threads occurs in many forms where it has not hitherto been described. Even in the Orthoptera, where the polarity is in general well marked, there is great variation in this respect in different species, and in some cases might easily be overlooked, as it seems to have been by McClung and Sutton.

Farmer and Moore (:05) emphasize the fact that there are always two distinct stages of the polar loops, which they have termed the first and second contraction figures. According to these authors, during the early growth period the spireme assumes the form of polar loops, which are always one-half as numerous as somatic chromosomes. "At the same time, the whole chromatic network contracts away from the nuclear membrane, this change producing the First Contraction figure. As time goes on the loops become not only increasingly chromatic but also opened out again, until the apparent polarisation is more or less completely lost and the nuclei present the well-known coarse spirem figure within the strands of which double beading or actual longitudinal fission is always more or less apparent. The coarse spirem figure often constitutes a prolonged phase, but it is in all cases ultimately succeeded by a short-lived and easily missed resumption on the part of the split chromatic thread-work of its earlier polarised arrangement; and this is followed by a strong Second Contraction and thickening of the individual loops."

It is very certain that in none of the Orthoptera which I have studied are there two contraction figures such as Farmer and Moore describe, nor is there at any time a continuous spireme. As I have previously described in detail, the original polarity persists until immediately previous to the time when the definitive tetrads are formed, and is then lost by the loops becoming detached from the nuclear membrane. In all cases the loops can still be seen to retain their connection with the pole even after they have opened out and assumed a peripheral position, while in some species the polarity is easily distinguished up to the time when the loops become free. Possibly the results of Farmer and Moore may be explained on the supposition that in Periplaneta, where the two contraction figures are especially well shown, the loops retain their connection with the nuclear wall until a later period than in most Orthoptera. If this connection should be retained until the loops begin to condense to form the tetrads, it would result in precisely such structures as are figured by these authors, except that between the two contraction figures the loss of polarity is only apparent.

It now remains to consider the method by which the reduction in the number of chromosomes during synapsis is brought about. According to most of the older accounts a continuous spireme was formed during the early growth stages, which later segmented into the reduced number of chromosomes. Montgomery (:00) was the first to clearly formulate the theory that the reduction is effected by the union of the chromosomes in pairs, and later (:01) strongly argued that one of each conjugating pair is derived from either parent. This theory of the conjugation in pairs of the maternal and paternal chromosomes during synapsis was supported by Sutton (:02, :03) and has now come to be generally accepted. However, there is still great diversity of opinion as to the manner in which the conjugation of the chromosomes takes place. In general two methods of chromosome conjugation have been described. According to one school of cytologists the chromosomes become united end to end, while the other school holds that they first become arranged parallel to each other and then unite side by side. In both types there is, according to the descriptions, great variation in the details for different species of animals, but that need not concern us here.

An end to end union of the chromosomes during synapsis has been described in amphibians by Montgomery (:03, :04), Moore and Embleton (:06); in selachians by Farmer and Moore (:05); in insects by Montgomery (:01, :05), Sutton (:02), Gross (:04), Farmer and Moore (:05), Nowlin (:06) and Stevens (:06[a]); in myriapods by Blackman (:05, :05[a], :07); in Peripatus by Montgomery (:00); in Allolobophora by Foot and Strobell (:05); and in Pedicellina by Dublin (:05). I believe that in the Orthoptera the evidence points strongly toward an end to end union, but not in the manner described by Sutton (:02). This author described and figured the members of the autosome pairs as becoming united at their ends nearest the spindle pole during the telophase of the last spermatogonial division. I have been unable to find any evidence of such a fusion at this time and believe Sutton was misled by an accidental approximation of the ends of the chromosomes, due to their being pulled toward a common point. That the conjugation takes place much later, is shown in Hippiscus and Melanoplus, where after the resting stage of the primary spermatocytes the chromatin collects in distinct masses, which probably represent univalent chromosomes. The actual union of the chromosomes probably occurs at about the time these chromatin masses become converted into spireme threads. This agrees with Montgomery (:05), who describes the end to end union in Syrbula as taking place during the early growth period. According to Stevens (:06^a) and Nowlin (:06) in various Coleoptera the numbers of polar loops is at first the same as the somatic number of autosomes, but later one end of each loop becomes free, the free ends then uniting in pairs.

On the other hand a side to side union of the chromosomes during synapsis has been described in mammals by von Winiwarter (:00, :02) and Schoenfeld (:01); in Amphibia by Janssens (:05), A. und K. E. Schreiner (:07); in fishes by Maréchal (:04), A. und K. E. Schreiner (:04, :05, :07); in insects by Otte (:06); in crustaceans by Lerat (:05); in gastropods by Bonnevie (:05, :06); in annelids by A. und K. E. Schreiner (:06, :06^a); in worms by Tretjakoff (:04) and Marcus (:06). The most detailed description of this type of conjugation has been given by A. und K. E. Schreiner (:04 to :07) in a series of papers on synapsis and maturation in various animals, in an avowed attempt to find a common type of these phenomena which will apply to all organisms. These authors find that in all the forms studied the number of polar loops is at first the same as the somatic number of chromosomes and only later is the number reduced to one half the somatic number, the reduction being accomplished by two loops becoming parallel and gradually fusing. They describe and figure the parallel approximation of the loops as taking place first near the pole where the loops are attached to the nuclear membrane, and gradually extending toward the opposite pole until the threads become connected throughout their entire length. The two components of the double thread thus formed then fuse into a single thread, which a little later splits along the line of fusion, so that the conjugants again become separated. The Schreiners find that the double thread is composed of a series of granules arranged in pairs, as I have described for the Orthoptera; but according to these authors one granule of each pair belongs to each conjugant, which means of course, that the chromosomes conjugate granule by granule, as the Schreiners (:07, p. 470) clearly state. "Der Prozess, den wir die parallele Konjugation der Chromosomen nennen, ist demnach nicht als eine Konjugation der Chromosomen als Ganzindividuen, sondern als *eine Konjugation der homologen Chromatineinheiten* aufzufassen, und dieser Konjugations typus ist eben in der Zusammensetzung der Chromosomen aus verschiedenen Einheiten bedingt."

I have already described at length my reasons for believing that such a method of conjugation does not occur in the Orthoptera, although Otte (:06) has described it for Locusta. I have occasionally seen the spireme threads lying parallel to each other in pairs near the distal pole, as described by the Schreiners, but believe this an accidental arrangement, which is more common near the pole, since in this region the threads are crowded more closely together. Moreover it seems hardly probable that the chromosomes should conjugate granule by granule.

Among the species in which the Schreiners have described the side by side union of the chromosomes is Salamandra, but both Montgomery (:03, :04) and Moore and Embleton (:06) have found an end to end union in closely related urodeles.

According to the Schreiners each of the polar loops later becomes converted into a tetrad in the following way: The two conjugants become widely separated, remaining connected only at one or both ends, to form loop- or ring-shaped elements, and at the same time a longitudinal split appears in each conjugant. This necessitates a very rapid shortening and thickening of the chromatic threads and the simultaneous appearance of a longitudinal split, but on both these points their figures are far from convincing.

It will be seen that the end result is the same as in Orthoptera, *i. e.* that the longitudinally split arms of the loops, or the opposite sides of the rings, represent the univalent components.

The types of bivalent chromosomes described by these authors in the prophase of the first division strikingly resemble those found in the Orthoptera.

4. The Maturation Divisions.

The extensive literature on this important stage in the development of the germ cells has been so often, and so thoroughly reviewed that it will be unnecessary for me to attempt an extended discussion here. The entire problem of maturation is at present in a very unsatisfactory state and there seems to be no prospect, for some time at least, of reducing the widely diverse accounts to a common basis, although the striking similarities in the shape and behavior of the bivalent elements in widely separated forms leads to the hope that in time this may be accomplished.

In any discussion of the subject the method of formation of the bivalent chromosomes must be taken into account and this has, of course, been done only within very recent years. For this reason the older accounts of the maturation division — such as those of Boveri ('87), Flemming ('88), Hertwig ('90), Brauer ('93), Meves ('96), von Lenhossék ('98), McGregor ('99), and Kingsbury (:02), in which both maturation divisions are held to be equational, since they are undoubtedly apparently longitudinal — are far from conclusive. As has been so often urged by Montgomery and others, the true interpretation of the maturation divisions must be sought in synapsis and the early prophase, and until these stages are fully elucidated it is impossible to arrive at conclusive results as to the significance of the two divisions. From this standpoint a number of recent writers have held that when there are two longitudinal divisions one is always reductional. In the Orthoptera we may have both a transverse and an *apparently* longitudinal division occurring in different chromosomes during the same mitosis, although when their previous history is taken into account it is evident that both divisions are fundamentally the same.

On the other hand, in the many instances where one transverse and one longitudinal division have been described, we may reasonably assume that the transverse division is probably reductional, even though the early stages in the formation of the bivalent chromosomes have not been fully worked out. However, even in such cases there is apparently room for doubt, since according to Struckman (:05) a transverse division may be equational.

Korschelt und Heider (:02) in their extensive review of the subject distinguish two types of maturation; the "eumitotic," where both divisions are equational and the "pseudomitotic," where one is reductional. The classical examples of the eumitotic type are the vertebrates and Ascaris, where, as described by a number of earlier investigators, two longitudinal divisions occur. But in the vertebrates Montgomery (:03, :04), A. und K. E. Schreiner (:04, :05, :07), Farmer and Moore (:05), and Janssens (:05) have found that one division is reductional, and in Ascaris Tretjakoff (:04) and Marcus (:06) have arrived at similar results, while Boveri (:04) has also argued for the probable occurrence of a reductional division in this form.

De Sinéty (:01) has described two longitudinal divisions in various Orthoptera and denies that either is a reduction division. His results have been criticised at length by McClung (:02) so that it is unnecessary to consider them in detail here. However, I am unable to agree in some cases with McClung's contentions. De Sinéty's interpretation of the maturation divisions appears to be based almost entirely on the larger chromosomes, where in some cases both divisions are *apparently* longitudinal. I believe that this author correctly described the division of the large ring- and loop-shaped chromosomes and that, contrary to McClung, the insertion of the spindle fibers may be "terminal," "subterminal" or "median" as de Sinéty maintained. This author also correctly described the halves of the rings as being pulled past each other during division. But I believe with McClung that de Sinéty was in error in maintaining that the ring- and loop-shaped chromosomes are formed by the opening out of the halves of a longitudinally split rod. In chromosomes of this type the inclosed space does not represent a longitudinal division, as de Sinéty believed, but separates the two univalent components. In short, in the case of the ring- and loop-shaped chromosomes my results agree with de Sinéty's as regards the division of these elements, but differ fundamentally with respect to their formation. In the case of the cross-shaped chromosomes I agree with McClung when he says: "Where the elements of one of these compound chromosomes intersect they lie in the same plane and are not superimposed upon each other as de Sinéty's theory demands and his figures represent."

Recently the eumitotic type of maturation has been revived by Bonnevie (:05, :06), who finds that in Enteroxenos, while the chromosomes conjugate side by side during synapsis, neither of the two succeeding divisions separates the conjugants but both are true equational divisions. However, judging from her figures, this species is a very unfavorable form in which to determine the plane of division.

In the case of the pseudomitotic type, Korschelt und Heider distinguish a "prereduction," where the first maturation division is reductional and a "postreduction," where the second division is the reducing one. The prereductional type has been described by a large number of writers among whom may be mentioned: Montgomery (:03, :04), A. und K. E. Schreiner (:04, :05, :07), Farmer and Moore (:05), and Janssens (:05) for vertebrates; Henking ('91), Paulmier ('99), Montgomery (:00, :01, :05 :06), Nichols (:02), Holmgren (:02) Farmer and Moore (:05), Lerat (:05), Wallace (:05) Stevens (:05 to :06[a]) Wilson (:05[a] to :06), Nowlin (:06) and Zweiger (:06) for arthropods; Korschelt ('95), Foot and Strobell (:05), and A. und K. E. Schreiner (:06, :06[a]) for annelids; Schockaert (:02) for Thysanzoon; Struckman (:05) for Strongylus; and Dublin (:05) for Pedicellina. Grégoire (:05), after an extensive review of the literature in the case of both animals and plants, concludes that this type will probably be found to be universal.

On the other hand the postreductional type has been described among others by vom Rath ('92, '95), McClung (:00, :02), Sutton (:02), Voinov (:03), Gross (:04), and Blackman (:05, :05[a], :06) for arthropods; by Griffin ('99) and Linville (:00) for molluscs; by Francotte ('97), Griffin ('99), and Tretjakoff (:04) for worms.

The results of most recent investigators, especially in vertebrates and arthropods, lend strong support to the view of Grégoire, that prereduction will be found to be the common if not universal type. Montgomery (:05) has strongly criticised the results of those who have described a postreduction and has shown that in many cases, at least, they are capable of a different interpretation. McClung (:00, :02) has described a postreduction in the Orthoptera (Acrididae and Locustidae), but, as Montgomery points out, in order to prove his point assumes a complicated series of changes to take place in the chromosomes during the late prophase. Apparently McClung has confused the rod- and cross-shaped chromosomes and considers that the former are always slightly later stages of the latter. Undoubtedly the cross-shaped elements do, as I have already described, become converted into rods during the late metaphase, but this is later than described by McClung and it by no means follows that the rods of the earlier stages are formed in this way.

Figure 62 (Plate 4) clearly shows several chromosomes which, previous to the formation of the equatorial plate, are arranged with their long axes parallel to the spindle axis. There is no evidence that at this early stage the chromosomes have begun to divide. Moreover, the rod-shaped elements can be distinguished throughout the entire prophase, as I have already described in detail.

McClung also lays great stress on the evidence afforded by the ring-shaped chromosomes, as the following quotation from him shows: "Reference was made on an earlier page to the conclusive evidence offered by the ring-figures with regard to the character of the first spermatocyte division. This, I think, cannot be disputed. The rings, with the point of cross-division to which the threads are attached indicated by a slight projection, come to lie in the equatorial plate. With the contraction of the fibers the halves of the rings separate more and more until, at the point of final separation, the resulting figure differs in no marked degree from that of the rod type." Contrary to McClung, I believe the evidence suggests strongly that the slight projection on the rings to which the spindle fibers are attached is not the point of cross-division but the point where the arms of the loops overlap. If the division of the rings is accomplished in the manner described by McClung, it is impossible to account for the crossed condition of the components at the point of separation, which undoubtedly occurs during the division of the ring-shaped chromosomes, but to which McClung makes no reference. However, in the forms upon which this author worked, it is difficult to demonstrate satisfactorily the sequence of the divisions, and I was myself for some time uncertain on this point. But in Stenobothrus I believe there can be little doubt that the arms of the large loops are separated during the first mitosis. McClung himself has later (:05) described a prereduction in certain chromosomes of various Orthoptera. In several of the Acrididae and one Locustid the monosome becomes attached to the end of a bivalent autosome during the prophase of the first division, and in such cases the autosome divides reductionally during the following division, although McClung still maintains that the remaining autosomes divide equationally. It is significant that in the autosome in which he finds a prereduction the free ends of the univalent components are so clearly marked that there can be no room for doubt on this point. In Mermiria this author finds that the element formed by the fusion of the monosome and a bivalent autosome end to end later unites with another bivalent autosome and that, "Upon the separation of the chromosomes in the metaphase the multiple chromosome is divided so that to one pole there goes a trivalent element and to the other a bivalent one, the difference in valence being due to the presence of the accessory chromosome in one daughter cell. There occurs here an entirely unique separation of chromosomes, for by means of it *entire tetrads pass into the second spermatocytes*." Such a division seems very improbable and requires much more conclusive proof than this author has been able to bring forward. A comparison of his Figure 12 with Figure 91 (Plate 6) of the present paper suggests that the "multiple chromosomes" of Mermiria may be capable of a different interpretation. This seems more probable when we remember that Mermiria and Stenobothrus are members of the same sub-family, the Tryxalinae. At another place in the same paper McClung refers to the unsymmetrical character of the daughter elements derived from the division of the multiple chromosome as follows: "Among the uncertainties in my mind concerning the behavior of the chromosomes in Mermiria, is one relating to the nature of the association of the chromosome into the multiple element of the first spermatocyte. The tetrads seem of the usual type, i. e. have simple chromosomes of equal size, but when the dyad divides it would appear as though there were some heterogeneity present, for in the anaphase one limb of the loop is longer than the other. This may be due to the formation of a multiple chromosome partly from the accessory chromosome; otherwise it means that the tetrad is not constituted of homologous simple chromosomes. Aside from this there seems to be nothing to contradict the view that the tetrads represent the union of homologous paternal and maternal elements." Such unsymmetrical loops always occur in Stenobothrus in the case of two of the larger bivalent chromosomes, as has been previously described in detail (Cf. Plate 6, Fig. 93). Sutton (:02) states that in Brachystola the first division is equational, but does not describe the process in detail. In Syrbula there is a prereduction, according to Montgomery (:05), and my results are a complete confirmation of his.

Voinov (:03) has described a post-reduction in Cybister, a coleopteron, but Holmgren (:02), Nowlin (:06), and Stevens (:06[a]) have found a prereduction in various forms of the same group. In the myriapods Blackman (:05, :05[a], :07) has described a postreduction, but the chromosomes in these forms appear to be very unfavorable for determining the sequence of the divisions, and I think are susceptible of either interpretation. This author lays much stress on the fact that in the early prophase the longitudinal cleavage appears before the transverse becomes evident and that it is but natural, therefore, to believe that the longitudinal division is the first to be completed in the two following mitoses. If the two divisions were of the same kind, no doubt this would be reasonable, but since they differ so fundamentally it is a question if the argument is valid.

Stevens (:03, :04) has found a prereduction in the spermatocytes of Sagitta and a postreduction in the oöcytes of the same form, but her figures are inconclusive.

It remains to consider the cases in which two transverse divisions have been described for the Orthoptera. Wilcox ('95) maintained that both divisions are transverse in Caloptenus (Melanoplus), but as his results have been generally discredited it is unnecessary to consider them here. Apparently Wilcox's fundamental error was in assuming that there are twelve chromosomes in the spermatogonia, whereas there are in reality twenty-three, as in the majority of the Acrididae. Recently Otte (:06) has described two transverse divisions in Locusta, but his results differ fundamentally from those of Wilcox, since he believes that neither is a reducing division in Weismann's sense. According to this author the chromosomes conjugate side by side during synapsis. Later the double threads thus formed shorten and thicken, while the free ends often approach each other and may fuse to form rings. "Da nun die Ringe durch Umbiegung eines Doppelfadens entstanden waren, so ist die 1. Reifungsteilung eine Querteilung des ursprünglichen Doppelfadens. Der Doppelfaden war durch parallele Conjugation zweier Einzelfäden entstanden. Diese beiden zu einem zweiwertigen Chromatinelement verbundenen Chromatinfäden wurden durch die Querteilung nicht auseinander gebracht, sondern quer halbiert." It is evident that in the case of the first division practically the only difference between Otte's results and my own is in regard to the formation of the bivalent chromosomes. In regard to the second division, however, our results are diametrically opposed. "Die Tochterchromosomen der 1. Reifungsteilung können bisweilen direkt in die 2. Reifungsteilung eintreten. Sie werden dañ entsprechend ihrer queren Einschnürung durchgeteilt. Gewöhnlich machen sie aber in den Spermatocyten II. Ordung mehr oder weniger starke Umwandlungen durch. Sie strecken sich und nehmen eine rauhe Oberfläche an. Es entstehen so wieder Schleifen, die sich zu Ringen umbilden können. Sie lassen ihre Doppelung, die Zusammensetzung aus zwei Fäden, oft deutlich erkennen. Auch typische Tetraden werden ausgebildet. Vor dem Eintritt in die 2. Reifungsteilung verdichten sich die Ringfiguren. In der 2. Reifungsteilung werden die Ringen wieder quer in zwei Halbringe geteilt. Auch die Tetraden werden ebenso wie in der 1. Reifungsteilung quer geteilt. In der Anaphase der 2. Reifungsteilung sieht man die Doppelung der Chromosomen noch recht deutlich."

In the second spermatogonia of Steiroxys none of the autosomes form rings, with the possible exception of the large element, and this, as my figures show, certainly divides longitudinally in the following division. In Steiroxys there are no tetrads in the second spermatocytes such as Otte describes, and in the anaphase of the second division I have been unable to find any evidence of a dyad structure in any of the chromosomes. In this connection I believe the evidence afforded by the large chromosomes is conclusive in showing that both divisions cannot be transverse. It might be argued with some degree of probabilty that both divisions of this chromosome are longitudinal, since both are undoubtedly apparently of this type, but I can find absolutely no evidence for two transverse divisions.

5. The Allosomes.

Inasmuch as extended discussions of these interesting elements have been given by a number of recent writers, notably Wilson (:**05**^{a} to :**06**) Montgomery (:**06**) and Gutherz (:**07**), it will be necessary here to consider them only with especial reference to the forms studied. The allosomes were first discovered in Pyrrhocoris by Henking ('**91**), who noted that one-half the spermatids received one more chromatic element than the others. They have since been described for a large number of forms but so far have been found only in the tracheates and arachnoids, with the possible exception of Sagitta. In insects they have been described by Montgomery ('98, Pentatoma, :01, :01[a], :06, Hemiptera, :05, Syrbula), Paulmier ('99, Anasa), McClung ('99, Xiphidium, :00, Hippiscus, :02[a], Locustidae), de Sinéty (:01, Orthoptera), Voinov (:03, Cybister) Baumgartner (:04, Gryllus), Gross (:04, Syromastes), Stevens (:05[a], Stenopelamtus, Blatella, Tenebrio), Montgomery (:05, Syrbula), Wilson (:05 to :06, Hemiptera), Nowlin (:06, Coptocycla), Stevens (:06[a], Coleoptera, Aphrophora, Cacoecia, Euvanessa), Zweiger (:06, Forficula), Otte (:06, Locusta), and Gutherz (:07, Gryllus, Pyrrhocoris). But Morgan (:06) and Stevens (:05, :06) have failed to find any allosomes in the aphids. The allosomes in myriapods have been described by Blackman (:05, Scolopendra) and Medes (:05, Scutigera); in arachnids by Wallace (:05, Agalena), Montgomery (:05, Lycosa) and Berry (:06, Epeira).

Until we learn more about the allosomes, they may for convenience be separated into two classes with subdivisions in each class (cf. Gutherz :07), although it is an open question whether in some cases the different types have any direct relation with each other. The different types of allosomes may be distinguished as follows:

A. Monosomes. These are allosomes which are unpaired in the spermatogonia and are usually more or less compact in the spermatocytes. They divide in only one of the two maturation divisions. These have been variously called accessory chromosomes, heterotropic chromosomes, odd chromosomes, etc.

The monosomes may be divided into two groups as follows: —

1. Monosomes which do not divide in the first maturation division, but do divide, probably equationally, in the second division. This type, with the possible exception of Syrbula (Montgomery :05), apparently occurs universally throughout the Orthoptera, and, with the exceptions noted, is the only type of allosome occurring in this group. Similar elements have been described in the Hemiptera by Wilson (:05[b], :06, Archimerus, Banasa), Montgomery (:06, Calocoris), and Stevens (:06[a], Aphrophora); in several Coleoptera by Stevens (:06[a]); in myriapods by Blackman (:05, Scolopendra) and Medes (:05, Scutigera); and in arachnids by Berry (:06, Epeira).

2. Monosomes which divide equationally during the first division, but fail to divide during the second. Such elements have so far been found only in the Hemiptera, where they have been described by

Henking ('91) and Montgomery (:01, :06) for Pyrrhocoris and by Wilson (:05^b) for a variety of Hemiptera.

A third kind of monosome, one which does not divide in either maturation division, has been described by Montgomery (:06) in Lygus, an hemipteran.

B. Diplosomes. These are allosomes which are paired in the spermatogonia and usually remain compact during the growth period of the primary spermatocyte. The diplosomes may be divided into three groups.

1. Diplosomes which typically are unequal in size, but may be equal. They may, or may not conjugate temporarily during the early growth period, but always enter the equatorial plate of the first division separately and there divide equationally. During the *second* division they divide reductionally, usually after a previous conjugation. These are the "idiochromosomes" of Wilson, and have been described in a large number of Hemiptera by Wilson (:05^a to :06) and Montgomery (:06).

2. Diplosomes, which, as in the first group, are typically unequal in size, but may be equal. They conjugate in the primary spermatocytes, usually during the early growth period, but do *not* separate before the *first* maturation division, where they divide reductionally. During the second division they divide equationally. These are the heterochromosomes of Miss Stevens; they have been described in a large number of Coleoptera by Stevens (:06^a) and in Coptocycla by Nowlin (:06); in the Lepidoptera by Stevens (:06^a, Cacaecia, Euvanessa); in the Orthoptera by Montgomery (:05) — this, however, needs confirmation; in Forficula by Zweiger (:06); and in arachnids by Montgomery (:05, Lycosa). Probably the larger pair of diplosomes described by Gross (:04) in Syromastes, as well as those in Tingis and Nabis (Montgomery :06), belong to this type.

3. Diplosomes which are usually much smaller than the other chromosomes and form a symmetrical pair in the spermatogonia. In most cases they do not conjugate until the prophase of the first division, where they divide reductionally. These are the m-chromosomes of Wilson and have been described for a variety of Hemiptera by Wilson (:05^b), Montgomery (:06) and Stevens (:06^a, Aphrophora). As Wilson points out, it is doubtful if they have any direct relation with the other allosomes.

Still other types of allosomes have been described in isolated cases, but as the accuracy of the results have been questioned, they may for our purposes be disregarded.

Various combinations of the allosomes in the same species occur in the Hemiptera, where they have been described by Wilson and Montgomery.

A glance at the above classification will show that among insects the first type of monosome (*A* 1) is characteristic of the Orthoptera, although it occurs in a few instances in the Hemiptera and Coleoptera. In the Orthoptera it has been found in all the forms investigated with the exception of Syrbula, Stenopelmatus, and Periplaneta. In Syrbula, one of the Acrididae, Montgomery (:05) found that the allosome of the spermatocytes is represented in the spermatogonia by two elements of equal size and is therefore a diplosome. During the early growth period the diplosomes become joined end to end to form a rod shaped element, which retains its compact structure and during the prophase of the first division becomes bent into a V. It divides reductionally in the first division and equationally in the second. These results of Montgomery's need confirmation, since all other observers who have traced the history of the allosome in the Orthoptera have found that it is unpaired in the spermatogonia and does not divide during the first division. Montgomery's Figure 33 is suggestive of an interpretation different from his, in that it shows a chromosome attached by mantle fibers to only one spindle pole, a condition that I have found to be realized in all the forms I have studied. Montgomery, however, believed that later this element divides. To quote:—"In a number of cases after nine of the chromosomes were arranged in the equator and some of these were beginning to divide (Fig. 33) one (*y*) had not yet taken up that position but lay nearer one spindle pole than the other. This was the case *e. g.* with four cells in exactly the same stage lying in the same section of one testicular follicle, and in all of these the isolated chromosome was of the same size and form, straight, and appearing to consist of two closely apposed arms. It may be that this is the heterochromosome, with which it agrees in general form and size, but this could not be definitely determined; ultimately it takes a position in the equator and divides with the others."

In Stenopelmatus Miss Stevens (:05[a]) has found that a conspicuous element, colored deeply with chromatin stains, appears in the early spermatocyte but is not represented in the spermatogonia. This peculiar element "first appears attached to an end of the spireme in the growth stage of the young spermatocytes, where it is much smaller than in later growth stages. It gradually increases in size, is a conspicuous element in the first maturation spindle, goes into one of each pair of spermatocytes of the second order, and then degenerates during the rest stage between the two maturation mitoses." In the present state of our knowledge it seems impossible to homologize this element with the monosome. In the case of Periplaneta Moore and Robinson (:05) have denied the presence of the monosome. The deeply staining body which can be seen applied to the nuclear membrane during the growth period they believe to be a plasmosome, which is extruded from the nucleus during the prophase of the first division. This seems in the highest degree improbable, since Stevens (:05^a) has found a typical monosome in the closely related germs Blatella. It seems probable that Moore and Robinson have confused the monosome and plasmosome. I have noticed that in many of the Acrididae during the late growth period, when the monosome stains less deeply and is usually greatly elongated, the plasmosome tends to take the chromatin stain and appears very similar to the monosome at an earlier stage. Baumgartner (:04) has noticed in Gryllus a similar tendency for the plasmosome to take the chromatin stain.

Recently Foot and Strobell (:07) have denied the presence of a monosome in Anasa, where it had been described by Wilson and by Montgomery. Foot and Strobell maintain that in the spermatogonia of Anasa there are twenty-two chromosomes, not twenty-one as held by Wilson and by Montgomery, and that the deeply staining element which is present in the spermatocytes during the growth period is simply a plasmosome, which is extruded from the nucleus in the prophase of the first division. They have failed to find any chromosome which does not divide in both divisions. However, Wilson (:07) asserts that reëxamination of his preparations has confirmed the accuracy of his previous results.

I think there can be no doubt that a monosome occurs in most of the Orthoptera. The cumulative evidence of a large number of investigators cannot be lightly put aside. I fail to see how in many cases a more conclusive demonstration could be desired. This is especially true of Steiroxys, where I have been able to follow the monosome continuously from the primary spermatogonia to a late stage in the metamorphosis of the spermatid.

In the spermatogonia of the Orthoptera the monosome is always one of the larger chromosomes and sometimes may be much larger than any of the autosomes. This is the case in Xiphidium (McClung, '99), Orphania (de Sinéty, :01) and Gryllus (de Sinéty, :01; Baumgartner, :04; Gutherz, :07). In the spermatocytes the monosome is always a conspicuous element, where, at least in the early growth stages, it forms a more or less compact body applied to the nuclear membrane and often in close connection with a plasmosome.

Among the most interesting phenomena exhibited by the monosome in the Orthoptera are the different forms which it assumes during the growth period. de Sinety (:01) first showed that in the Phasmidae the monosome becomes much elongated during this period and in some cases may be in direct connection with the spireme, while McClung (:02) and Otte (:06) have shown that in the Locustidae the monosome becomes converted into a long coiled thread. According to Otte this thread shortens and thickens to form a loop, which by the apposition of its arms becomes converted into a longitudinally double rod. My own observations have shown that in the Acrididae the monosome has a somewhat similar history, although there is considerable variation in the different subfamilies. In Steiroxys, however, the monosome apparently remains more compact than in most of the Locustidae. The entire history of this element is strikingly like that of the autosomes, but with this difference: — in the monosome the different autosome stages have been suppressed to a very large extent. It is only necessary to add that it forms a modified spireme and is attached to the distal pole in the same manner as the autosomes, although in the monosome the spireme is found at a considerably later period.

In the Orthoptera, as in other insects, the monosome often shows a distinct bipartite structure. This is especially true of Stenobothrus and Melanoplus, where, as I have shown, during a large part of the growth period the monosome is composed of two distinct and dissimilar portions.

McClung (:05) has described some peculiar relations between the autosomes and monosome in certain Orthoptera. In several species the monosome becomes attached to a bivalent (in Mermiria quadrivalent) autosome during the prophase of the first division and this association persists throughout the maturation period. A similar condition was noticed by de Sinéty in the phasmid Leptynia. I have been unable to find anything of the kind in any of the Orthoptera studied. However, a comparison of McClung's figures with some of the stages of the monosome in Stenobothrus suggest that possibly in some cases he has mistaken the more granular component of the monosome for an autosome.

Regarding the relation of the monosome of the Orthoptera to the allosomes of other arthropods, it is probably directly comparable with the monosomes of other forms, whether they divide in the first or second division, since the time of division would seem to be a character of secondary importance. In regard to its connection with the diplosomes, we are apparently on less certain ground. Wilson has suggested that the monosomes simply result from a further modification of the unsymmetrically paired diplosomes, the smaller component of the pair having been lost altogether. On this view it seems impossible to account for their frequent bipartite structure, which has led Montgomery (:05) to argue that the monosome may be formed by the permanent fusion of a diplosome pair. However, he later (:06) pointed out that there are difficulties in the way of such a view, since Stevens (:05, :06[a]) and Wilson (:05 to :06) have shown that where there is a monosome in the male there is a symmetrical pair of chromosomes in the female, and where in the male there is a pair of diplosomes of unequal volume these are represented in the female by a pair of equal volume. This of course means that in species where the spermatozoa are dimorphic, one-half containing a monosome and the other half lacking this element, the mature eggs are not dimorphic, for each contains the equivalent of a monosome. Similarly, in cases where one-half the spermatozoa contain the large, the other half the small diplosome, all the mature eggs contain a chromosome corresponding to the large diplosome. From this both Stevens and Wilson conclude that an egg when fertilized with a spermatozoön containing the monosome or large diplosome develops into a female, but when fertilized with a spermatozoön lacking the monosome or large diplosome develops into a male. My own observations in Hippiscus indicate that a similar condition exists in the Orthoptera. It will be remembered that in the oögonia of this species there is a symmetrical pair of chromosomes which correspond to the monosome in the spermatogonia. This being the case, it is difficult to explain the monosome as originating by the permanent fusion of two formerly distinct elements, for on this view it would seem that the number of chromosomes in the female should be one less than in the male, instead of one more as is actually the case. Unfortunately we know nothing concerning the behavior of the allosomes or their equivalents during the maturation and fertilization of the egg and until these stages have been fully elucidated we can scarcely hope to arrive at any adequate explanation of their origin.

A number of theories have been developed regarding the function of the allosomes, but so far with very indifferent success. Paulmier ('99) and Montgomery (:01) have suggested that they are degenerating chromosomes, but Montgomery has recently (:06) receded from his former position, holding simply that their function must be very different from that of the autosomes, as indicated by their structure, without attempting any specific statement. McClung (:01), Stevens (:05[a], :06[a]) and Wilson (:06) have pointed out the possible significance of the allosomes in sex determination, so that it is unnecessary to take up this subject, especially since in the present state of our knowledge it can only lead to fruitless theorizing. I think it must be conceded that we have at present no satisfactory explanation of either the origin or function of the allosomes, and until we know more about these interesting elements, especially during oögenesis and fertilization it is idle to speculate further on the subject.

6. The Individuality of the Chromosomes.

The theory of the individuality of the chromosomes, which was first formulated by Rabl ('85) and later ardently advocated by Boveri, has in recent years received strong support, especially from writers on insect spermatogenesis. The constant recurrence in successive generations of the autosome pairs, which are so clearly marked in these forms, is very difficult to explain on any other view. When we consider that each species is characterized by a fixed number of symmetrically paired autosomes which possess definite form and size relations, it is almost impossible to escape the conclusion that we are dealing with distinct morphological entities. This is perhaps nowhere shown to better advantage than in the aphids, where Stevens (:05, :06, p. 15) has shown that: "In every one of the twenty-four species examined some or all of the chromosomes possess characteristics which distinguish them from their fellows, and these peculiarities persist throughout all the generations. In every species where it has been possible to study and compare the germ cells of the parthenogenetic and sexual generations the single series of the maturating sexual germ cells has been found to be exactly duplicated in the double series of the parthenogenetic egg, the segmenting winter egg and the spermatocytes before reduction."

The Orthoptera also furnish strong evidence for the individuality of the chromosomes especially in the case of Stenobothrus and Steiroxys, where, as previously described, the autosomes vary in their method of attachment to the mantle fibers. In Stenobothrus five of the eight autosome pairs always have the mantle fibers attached to one end. In two of the remaining three pairs the mantle fibers are inserted nearer one end than the other, while in the third pair the insertion is at the center. In Steiroxys the insertion is at the end in all except one pair of autosomes, while in that pair it is always near the middle.

Although the chromosomes seem to lose their identity during the resting stage, this must be only apparent, since in the prophase they reappear with the same orientation as before. Similar conditions have been described by Rabl ('85) in Salamandra and Boveri ('88) in Ascaris. Recently Otte (:06) has found that in the spermatogonia of Locusta each chromosome remains in a distinct vacuole during the resting stage, while Farmer and Moore (:05) have found in Periplaneta, and Moore and Embleton (:06) in Triton, that although a common nuclear membrane is formed in the resting spermatogonia, the individual chromosomes can still be distinguished. Of interest in this connection are the results of Maréchal (:04, :05), who finds that in the oöcytes of certain fishes the chromosomes gradually become less distinct during the growth stage owing to the fact that the chromatin travels out along fine threads, while the axis still remains distinct as a somewhat more deeply staining mass.

Of especial interest are the results of Moenkhaus (:04), who was able in hybrids between Fundulus and Menidia to distinguish the chromosomes of either parent up to the late cleavage stage. Even more striking results have been obtained in plant hybrids.

Evidence of still greater weight is furnished by the occurrence of bivalent chromosomes of constant form in the spermatocytes. Baumgartner (:04) has found that in Gryllus autosomes in the form of rings, crosses and rods constantly occur and that there is probably a fixed number of elements of each type, while Nichols (:06) has described similar conditions in the spermatocytes of Oniscus. Recently Moore and Arnold (:06) have investigated a number of animals from this standpoint and find that in the spermatocytes of each form studied several types of bivalent chromosomes ("gemini") occur, and that there are always a fixed number of each type, although the number and form of the different types varies widely in different species. They conclude: "What appears to us of first importance is the recognition of the actual existence of permanent structural types in the gemini of different forms. Secondly, it would appear that in any particular form the number of gemini of each type have a constant numerical relation to each other. Thirdly, so far as the investigation has at present gone certain types of gemini appear to be common in all the widely sundered forms."

In Dissosteira there is a similar constancy in the different types of bivalent autosomes, and although in the other forms a similar detailed study of the autosomes was not made, yet where, as in Stenobothrus and Steiroxys, there are certain easily distinguishable elements, these always occur with constant regularity. Moreover, in addition to their constant form, certain chromosomes have a definite mode of attachment to the mantle fibers, which persists throughout all the generations.

In the case of the allosomes, as has been urged by a number of writers, we have very strong evidence in favor of the individuality of the chromosome. Certainly there can be no doubt that in many cases at least these elements retain their individuality from one generation to another. This is especially true of Steiroxys, where the monosome can be followed continuously from the primary spermatogonia to the spermatid. Furthermore in the case of Arphia we have in one individual two monosomes persisting throughout the spermatogenic cycle, and this abnormality is constant for all the testicular elements. Similar phenomena have been described by Stevens (:06[a]) and Zweiger (:06).

But this morphological differentiation of individual chromosomes must mean a corresponding physiological differentiation, as was first clearly brought out by Boveri and Sutton. Boveri's (:02) remarkable experiments, which are too well known to require discussion here, have led him (:04) to conclude: "Somit bleibt keine andere Annahme übrig, als dass die Variationen, die wir in der Entwicklung dispermer Keime angetroffen haben, auf verschiedener Kombination von Chromosomen beruht, und dies heisst nichts anderes, als dass die einzelnen Chromosomen verschiedene Qualitäten besitzen müssen." This is entirely in accord with the morphological differences in form and volume, for, as Montgomery (:06) has pointed out, chromosomes of different size cannot have the same physiological value but must have activities differing at least in amount. Moreover, the constant difference in form which has been shown to occur can be explained only on the basis of a physiological difference of which the form is the expression. Then, too, we have the evidence of the allosomes, whose functions as indicated by their very different form and behavior must be quite unlike that of the autosomes. Further, there seems to be little question that, as first argued by Sutton (:03), we are justified in concluding that the paternal and maternal components of each autosome pair are practically alike physiologically as well as morphologically. Such a physiological similarity would explain the intimate relations which are found to exist at all times between the components of each pair as well as their conjugation during synapsis. This would also indicate, as first suggested by Sutton (:02) and later elaborated by Boveri (:04), that possibly the conjugation of the chromosomes during synapsis is not so much to allow for an intermingling of the substance of the conjugants as to afford a simple means of insuring that two homologous chromosomes shall not enter the same spermatid or mature egg.

Finally I may point out that the behavior of the chromosomes in the Orthoptera during spermatogenesis is fully in accord with the Mendelian law of alternative inheritance, as has been pointed out by Sutton (:03).

7. The Metamorphosis of the Spermatid.

One of the most surprising results has been to find such a great difference between the Acrididae and the Locustidae in the metamorphosis of the spermatids. The fact that the spermatids of two such closely related families differ so markedly during metamorphosis would seem to indicate that the details of the process can have no fundamental significance. It does not seem best therefore to attempt any wide comparisons, since they would appear to be of doubtful value. One of the most common and characteristic structures of the spermatids is the so called Nebenkern, yet in the Acrididae its history is very different from that of a similar element in Steiroxys. But it may be argued that the Nebenkerne in the two cases are not homologous structures. Possibly — very probably — they are not, but the point which I wish to emphasize is that in the early spermatids we have in both cases structures which *appear* the same, and no one examining the spermatids at this time would hesitate, I think, to conclude that they are similar structures. Yet it is very certain that later the Nebenkern in the Acrididae becomes converted into a sheath surrounding the axial filament, while in Steiroxys after a complicated metamorphosis, it forms the acrosome. Unfortunately I have not been able to determine the origin of the Nebenkern in Steiroxys and it may be that Steiroxys differs in this respect from the Acrididae. However, I hope later by comparison with other forms to clear up this point. Meanwhile, the metamorphosis of the spermatids in Locusta as described by Otte (:06[a]) leads to some interesting suggestions. This author found that in Locusta the mitochondrion forms in the spermatocytes a distinct body, which usually shows an annular differentiation. During the prophase of the first division it divides into a number of small bodies, which are irregularly distributed to the daughter cells. In the spermatids most of the mitochondrion collects into a compact "Mitochondrienkörper" (Nebenkern). Meanwhile the interzonal filaments, with possibly part of the mitochondrion, become converted into a rounded "Idiozom." Later the "Mitochondrienkörper" elongates to form a sheath around the axial filament, while the idiozome becomes applied to the anterior end of the nucleus and forms an anchor-shaped acrosome. In this brief paper Otte does not describe the formation of the acrosome in detail and, as he gives no figures, it is impossible to determine whether the process is similar in the two cases, but his reference to the differential staining of the acrosome would suggest that the process is much the same as in Steiroxys. "Die färbbare Substanz ordnet sich auf den verschiedenen Stadien der Ausbildung verschieden im Spitzenstück an, so dass nach Heidenhainscher Färbung oft recht eigenartige Differenzierungen im Spitzenstück entstehen. Gegen die Vollendung der Ausbildung des Spermatozoons verteilt sich die färbbare Substanz vollkommen gleichmässig über das Spitzenstück; nur die vorderste Spitze erscheint frei davon" (Otte, :06^{a}, p. 753).

Apparently the conspicuous spherical body in the spermatids of Steiroxys, which I have called Nebenkern, is comparable to the idiozome of Otte. This being the case, the small, irregularly shaped body which is applied to the nuclear membrane near the Nebenkern in the young spermatids would seem to correspond to the "Mitochondrienkörper," although there is nothing in its appearance to suggest such a comparison. Following out this line of comparison still further, it would appear that the Nebenkern in the Acrididae is not comparable with the structure to which I have applied that term in Steiroxys, but rather to the small inconspicuous body which lies by the side of the nucleus, although in appearance the two are very unlike. If this comparison is well founded, then there is nothing in the spermatids of the Acrididae corresponding to the body which I have called the Nebenkern in Steiroxys, and yet, with the exception of the nucleus, it is the most conspicuous element in the spermatid. On the other hand, the small indistinct body which lies beside the nucleus in Steiroxys has the position and to a less extent the appearance of the body which has been described as the acrosome in the spermatids of various insects and traced into the anterior end of the spermatozoön. Such structures have been described by Henking ('91, Pyrrhocoris), Paulmier ('99, Anasa), Baumgartner (:02, Gryllus), Stevens (:05^{a}, Stenopelmatus).

Regarding the origin of the Nebenkern in insects, there appear to be in general two views. La Vallette St. George ('86), Henking ('91), Paulmier ('99), Meves (:00, :02), Holmgren (:02) and Zweiger (:06) have found that it is derived chiefly from the mitochondrion, while

Platner ('89), Wilcox ('95) Erlanger ('97), Baumgartner (:02) and Munson (:06) have maintained that it is formed almost entirely from the interzonal filaments. In Dissosteira I think there can be no question that the Nebenkern is not formed, directly at least, from the interzonal filaments, since they can be plainly distinguished after the formation of the Nebenkern is well advanced. It is of course possible that as the interzonal filaments disappear the material derived from their disintegration may aid in the formation of the Nebenkern, but this seems scarcely probable. On the other hand there is good evidence that it is formed from the mitochondrion.

V. Summary.

1. In all the forms studied there is a single apical cell of characteristic appearance at the distal end of each follicle.
2. The primary spermatogonia surround, and are in contact with, the apical cell.
3. The secondary spermatogonia are inclosed within a membrane formed by connective-tissue cells, the whole constituting a spermatocyst.
4. All the spermatogonia in each cyst are the direct descendants of a single primary spermatogonium, which became surrounded by one or more connective-tissue cells, and are, with rare exceptions, in practically the same stage of development.
5. The resting nuclei of the spermatogonia are irregular in shape and show a marked depression on the side adjacent to the greatest amount of cytoplasm.
6. The autosomes of the spermatogonia vary greatly in size, and can be readily arranged in symmetrical pairs, which usually lie close together, and show constant and characteristic differences in both form and size.
7. A monosome is always present.
8. The oögonia contain one more chromosome than the spermatogonia, there being a symmetrical pair in place of the monosome.
9. In the resting spermatogonia of Steiroxys trilineata the monosome is inclosed within a separate vesicle.
10. During the telophase of the last spermatogonial division the monosome, which is inclosed within a distinct vesicle, retains its compact form and often shows a more or less distinct bipartite structure.
11. The first stage of the primary spermatocyte is characterized by the chromatin being evenly distributed through the nucleus in a finely granular condition.

12. Later the chromatin collects in more or less definite masses, which, in favorable cases (Chortophaga, Melanoplus), can be seen to be of approximately the same number as the autosomes of spermatogonia.

13. Each chromatic mass later becomes converted into a single spireme thread, composed of a single series of chromatin granules connected by linin.

14. The spireme threads become converted into loops having a polar arrangement, each loop being composed of two homologous autosomes joined end to end.

15. The polar loops later show a more or less distinct longitudinal split. This split, however, does not extend to the linin, but is produced by each chromatin granule dividing into two equal parts.

16. Later the longitudinal split becomes temporarily indistinct, and may entirely disappear, while the loops open out and assume a peripheral position.

17. In the early growth period the monosome becomes inclosed within the nucleus, where it forms a somewhat flattened, deeply staining, often vacuolated element closely applied to the nuclear membrane.

18. During the later growth period the monosome goes through a complicated development, which, to a certain extent, is comparable to that undergone by the autosomes during the same period. In Stenobothrus and Melanoplus the monosome divides into two dissimilar parts, which can be distinguished up to a late stage in the prophase of the first maturation division.

19. Each polar loop of the growth period develops into a definitive tetrad during the prophase of the first division.

20. The bivalent autosomes, or tetrads, show the same size relations as the autosome pairs of the spermatogonia, and are evidently formed by the conjugation of the components of each pair.

21. In addition to the difference in volume, the bivalent autosomes show constant and characteristic differences in form. In general several more or less distinct morphological types can be distinguished, and the members of each type appear to bear a constant numerical relationship to each other.

22. The first maturation division is reductional, separating homologous chromosomes, which united during synapsis.

23. The second division is equational.

24. Individual chromosomes differ in regard to the point of insertion of the spindle fibers during mitosis, but for each chromosome this point is constant throughout the spermatogonial and spermatocyte divisions.

25. The monosome does not divide in the first division but divides longitudinally and probably equationally in the second.

26. The spermatids are dimorphic, one-half containing a monosome while the other half lack this element.

27. The monosome remains compact for some time within the nucleus of the spermatids, but later breaks up into fine granules in the same manner as the autosomes.

28. In Dissosteira carolina the Nebenkern is not derived directly from the remains of the spindle fibers but is probably formed chiefly from the mitochondrion.

29. In Dissosteira carolina the axial filament is from the first connected with a distinct centrosome, which is applied to the exterior of the nuclear membrane. As the spermatid elongates the Nebenkern becomes converted into an envelope surrounding the axial filament.

30. In Dissosteira carolina the head of the mature spermatozoön is formed entirely from the nucleus; the centrosome forms the greater part of the small middle-piece; while the tail is composed of a central fiber, derived from the axial filament and Nebenkern, surrounded by a cytoplasmic envelope.

31. In Steiroxys trilineata the axial filament is apparently not at first connected with a centrosome, which appears in the usual position only when the axial filament is well developed. The axial filament is from the first surrounded by an envelope of doubtful origin denser than the surrounding cytoplasm. The Nebenkern migrates around the nucleus and becomes applied to its anterior end.

32. In Steiroxys trilineata the head of the mature spermatozoön is formed from the nucleus and the Nebenkern, the latter developing into the acrosome; the middle piece is formed chiefly from the centrosome, which divides into four parts; while the tail is composed of the central fiber, derived in part from the axial filament, surrounded by a cytoplasmic envelope.

33. In no case is there evidence that the monosome is extruded from the nucleus during metamorphosis or that the spermatids degenerate, except in rare instances. Therefore the mature spermatozoa must be dimorphic with respect to their chromatin content, although there are no visible differences either in form or volume between the two types.

34. Throughout the entire history of the germ cells there is strong evidence for the individuality of the chromosomes.

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Explanation of Plates.

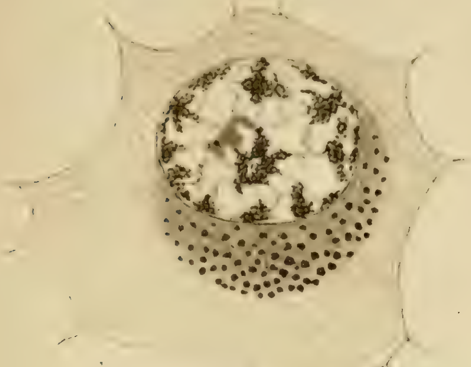
All drawings have been made at the level of the base of the microscope with the aid of a camera lucida. The combination used, Leitz $\frac{1}{12}$ inch objective and No. 4 ocular, was the same in all cases. All figures with the exception of Plate 4 are magnified 1450 diameters. The figures in Plate 4 have been reduced $\frac{1}{3}$, giving a final magnification of 966 diameters.

PLATE 1.

FIG. 1. Apical cell of *Dissosteira carolina.*
FIG. 2. Apical cell of *Stenobothrus curtipennis.*
FIG. 3. Apical cell of *Melanoplus femoratus.*
FIG. 4. Apical cell of *Steiroxys trilineata.*

Figs. 5–15. *Dissosteira carolina.*

FIG. 5. Resting stage of primary spermatogonium.
FIG. 6. Resting stage of secondary spermatogonium.
FIGS. 7, 8. Prophase of secondary spermatogonia.
FIG. 9, 10. Same stage as Figs. 7 and 8, but the sections are cut in such a way as to show only one side of the nucleus.
FIG. 11. Late prophase of secondary spermatogonium.
FIG. 12. Polar view, metaphase of secondary spermatogonium.
FIG. 13. Side view of the same stage as Fig. 12.
FIG. 14. Anaphase of secondary spermatogonium.
FIG. 15. Telophase of secondary spermatogonium.
FIG. 16. Prophase of secondary spermatogonium of *Steiroxys trilineata.* The monosome is shown inclosed in a separate vesicle.
FIG. 17. Prophase, secondary spermatogonium of *Stenobothrus curtipennis.*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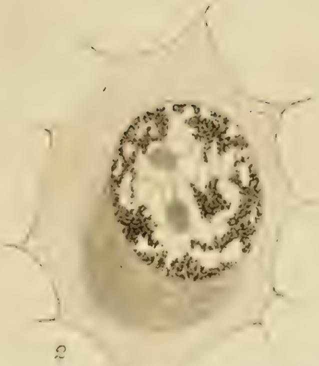
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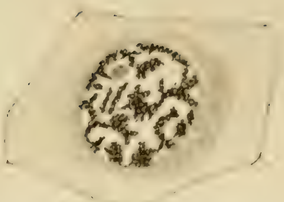
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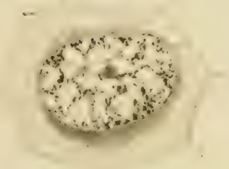
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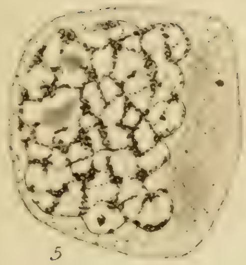
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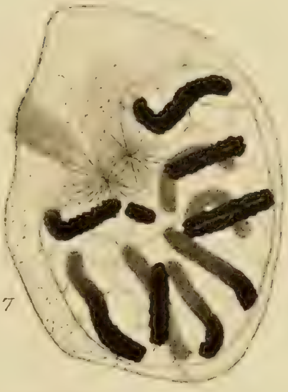
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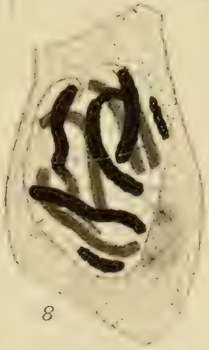
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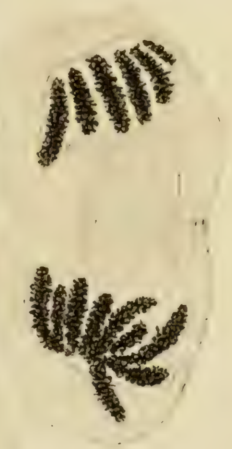
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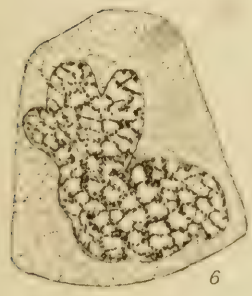
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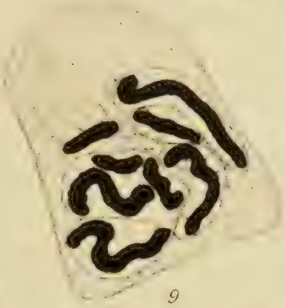
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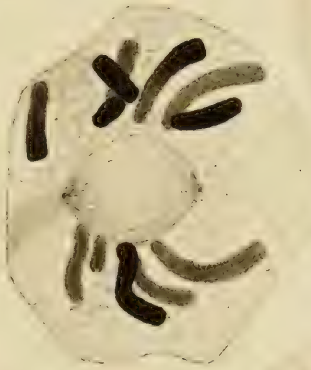
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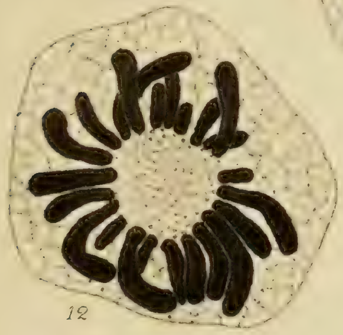
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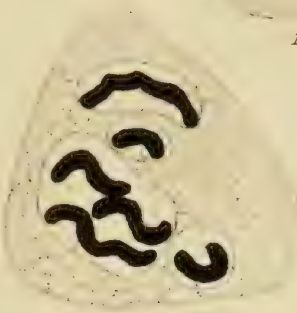
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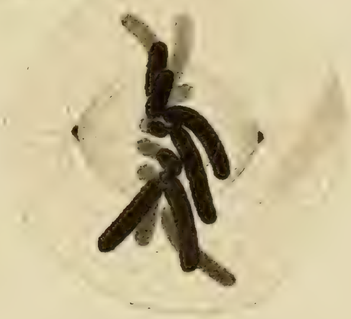
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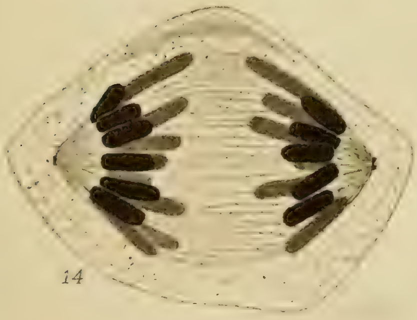
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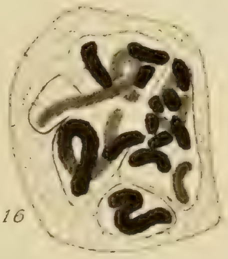
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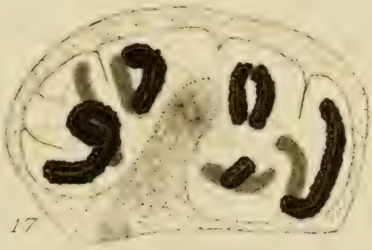
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H. S. D. del.

PLATE 2.

Figs. 18–20. *Steiroxys trilineata.*

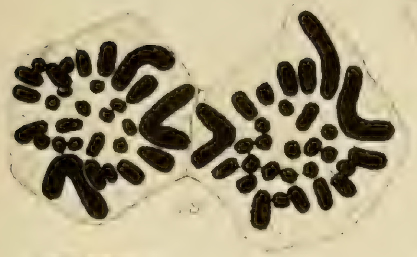
Fig. 18. Polar view, metaphase of secondary spermatogonia.
Fig. 19. Resting stage of secondary spermatogonia. The monosome lies outside the nucleus in a separate vesicle.
Fig. 20. Metaphase of secondary spermatogonia.
Fig. 21. Prophase of secondary spermatogonia in *Stenobothrus curtipennis.*

Figs. 22–26. *Dissosteira carolina.*

Fig. 22. Cross section of chromosomes during telophase of last spermatogonial division. The monosome is compact and inclosed in a separate vesicle.
Fig. 23. Telophase of last spermatogonial division. The monosome shows a distinctly bipartite structure.
Figs. 24, 25. Late telophase of last spermatogonial division. Owing to imperfect fixation the chromatin is shrunken away from the nuclear wall. The monosome is still inclosed in a separate vesicle.
Fig. 26. Stage *a,* primary spermatocyte. The monosome is shown above.
Fig. 27. Stage *a,* primary spermatocyte of *Arphia tenebrosa.* Two monosomes are present.

Figs. 28–34. *Dissosteira carolina.*

Fig. 28. Stage *b,* primary spermatocyte.
Fig. 29. Stage *c,* primary spermatocyte. The monosome now lies within the nucleus.
Fig. 30. Stage *c,* a little later than Fig. 29.
Figs. 31, 32. Stage *d,* primary spermatocyte.
Fig. 33. Stage *e,* distal pole of nucleus of primary spermatocyte.
Fig. 34. Stage *e,* primary spermatocyte.
Fig. 35. Stage *e,* distal pole of nucleus of *Arphia tenebrosa.* The monosome is seen below in the figure.
Fig. 36. Stage *e,* cross section of polar loops near distal pole.



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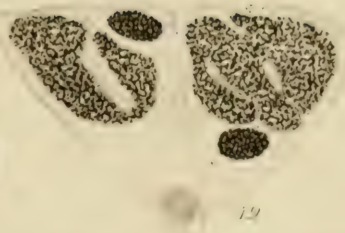

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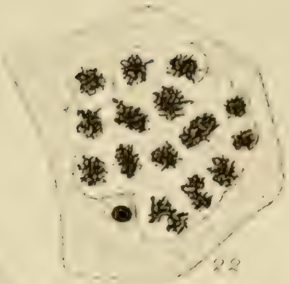

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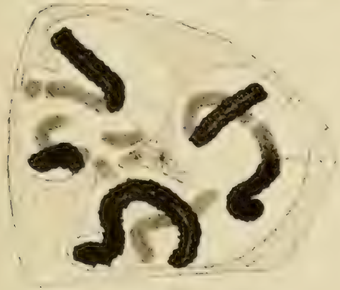

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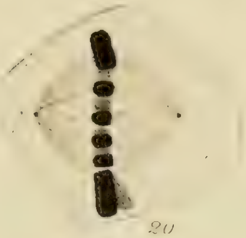

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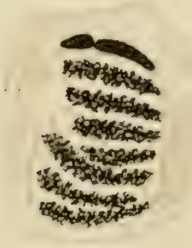

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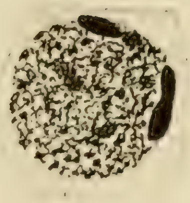

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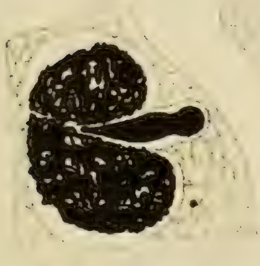

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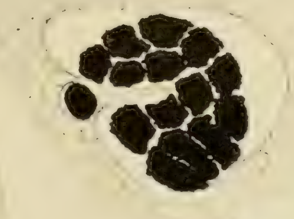

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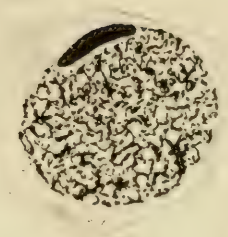

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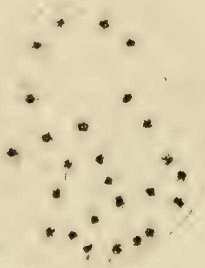

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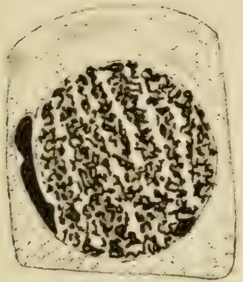

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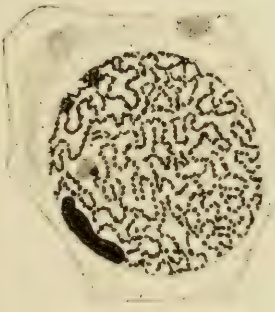

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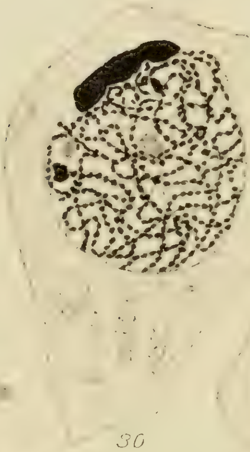

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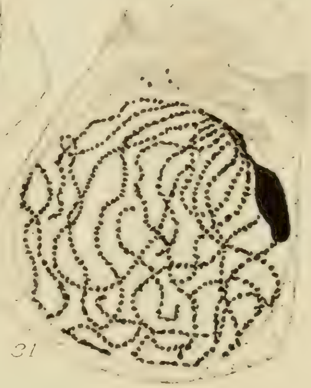

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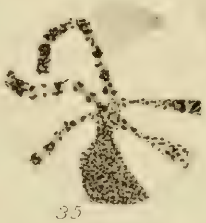

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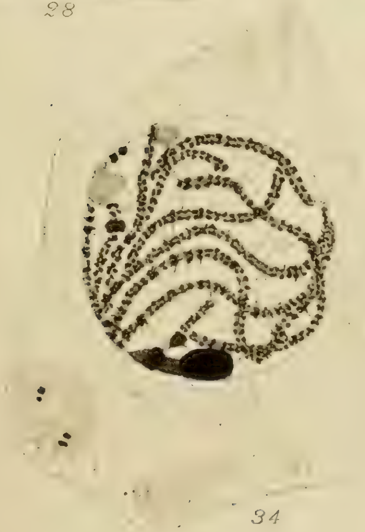

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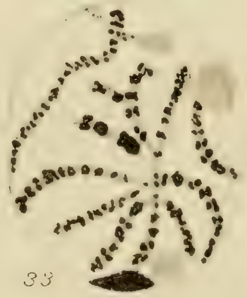

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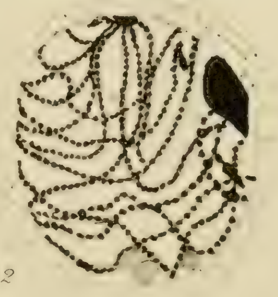


32

J S D del

PLATE 3.

FIG. 37. Stage *d*, primary spermatocyte of *Arphia tenebrosa*. Two monosomes are present.

FIGS. 38–40. Nuclei of primary spermatocytes of *Hippiscus tuberculatus* showing successive stages in the splitting of the spireme threads.

Figs. 41–44. *Chortophaga viridifasciata.*

FIGS. 41, 42. Stage *b*, primary spermatocytes. The plane of Fig. 42 is perpendicular to that of Fig. 41.

FIGS. 43, 44. Nuclei slightly older than those of Figs 41, 42, showing development of spireme threads from chromatic masses.

FIG. 45. Stage *b*, primary spermatocyte of *Melanoplus femoratum.*

FIG. 46. Stage *f*, primary spermatocyte of *Dissosteira carolina*. The monosome is seen below at the right.

Figs. 47–52. *Stenobothrus curtipennis.*

FIG. 47. Stage *a*, primary spermatocyte. The monosome is inclosed in a separate vesicle.

FIG. 48. Stage *e*, primary spermatocyte.

FIGS. 49–50. Stage *e*, nuclei of primary spermatocyte. In Figure 49 the monosome lies below and at some distance from the distal pole, while in Figure 50 it lies close to the pole.

FIG. 51. Stage *e*, cross section of polar loops and monosome near the distal pole.

FIG. 52. Stage *f*, primary spermatocyte. The bipartite structure of the monosome is well shown.

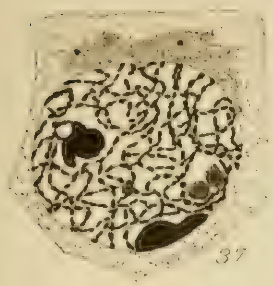
Figs. 53–56. *Steiroxys trilineata.*

FIG. 53. Stage *a*, primary spermatocyte. Owing to imperfect fixation the chromatin is shrunken away from the nuclear wall. The monosome lies outside the nucleus.

FIG. 54. Same stage as Figure 53, but the fixation of the cell is good.

FIG. 55. Stage *c*, primary spermatocyte. The monosome has become applied to the nuclear membrane.

FIG. 56. Stage *d*, primary spermatocyte. The monosome now lies within the nucleus. The mitochondrion body is seen in the cytoplasm above the nucleus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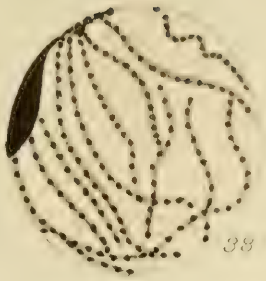
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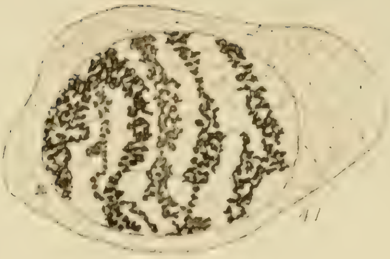
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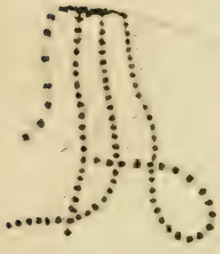
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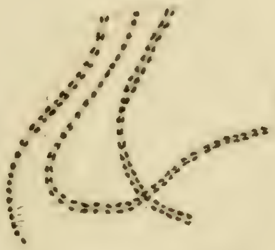
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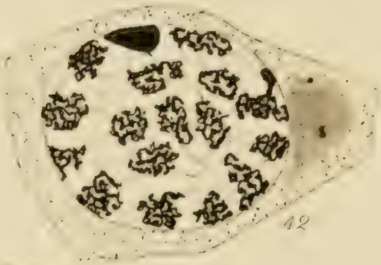
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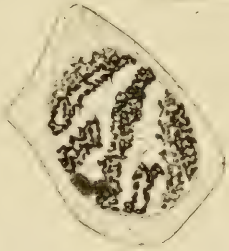
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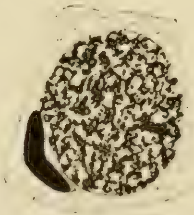
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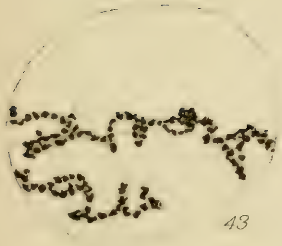
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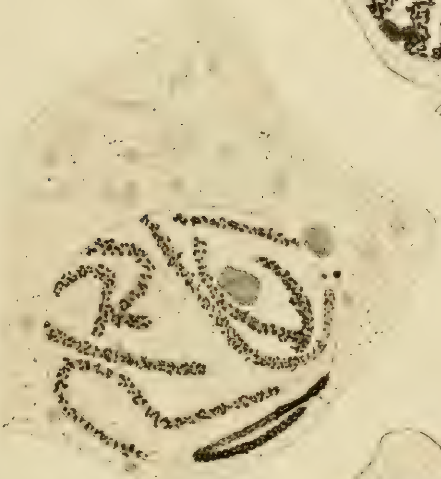
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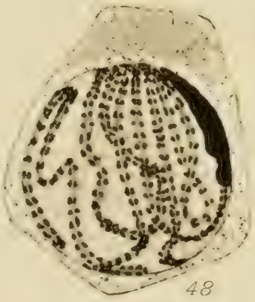
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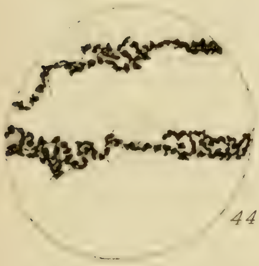
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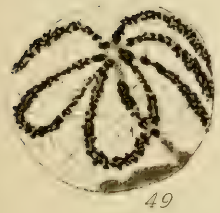
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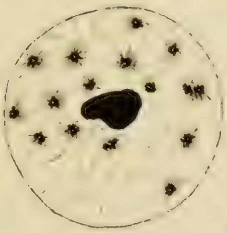
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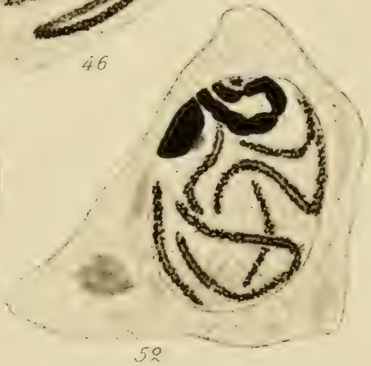
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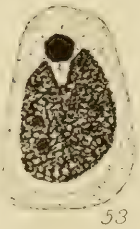
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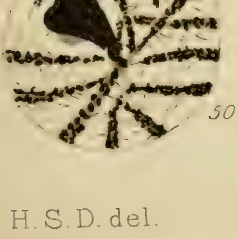
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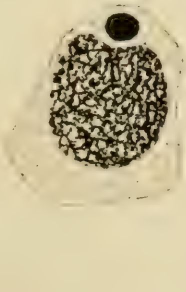
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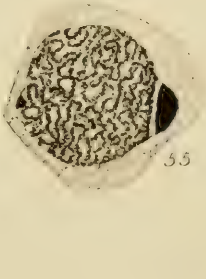
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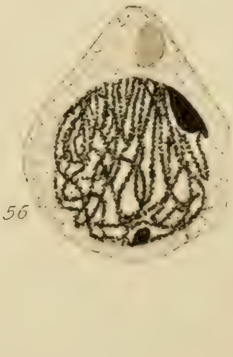

56

H.S.D. del.

PLATE 4.

All figures magnified 966 diameters.

Figs. 57–63. *Dissosteira carolina.*

Figs. 57, 58. Stage *g*, primary spermatocyte. The loop-shaped monosome is shown in both cases.

Figs. 59–60. Stage *h*, primary spermatocytes. The tetrads are already formed. The rod-shaped monosome is seen in each cell. The irregular deeply staining body in the upper part of the nucleus in Figure 59 is a plasmosome.

Fig. 61. Stage *i*, primary spermatocyte. The monosome is seen at the right applied to the nuclear membrane.

Fig. 62. Late prophase of first maturation division.

Fig. 63. Metaphase of first maturation division. The rough rod-shaped element lying at an angle to the spindle axis is the monosome.

Fig. 64. Metaphase, first division of *Arphia tenebrosa.* A monosome lies near each spindle pole.

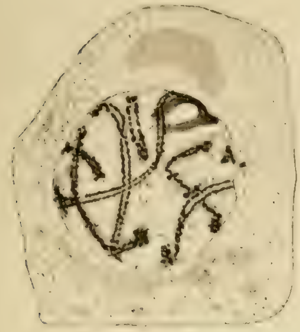
Figs. 65–68. *Dissosteira carolina.*

Fig. 65. Early anaphase of first division. The monosome lies near the left spindle pole.

Fig. 66. Late anaphase of first division.

Fig. 67. Group of cells from the distal end of the follicle. The apical cell is seen below and at the right — the one nearest the numerals "67." Surrounding the apical cell above and at the left are three primary spermatogonia in different stages. Above and at the left of the primary spermatogonia are four connective-tissue cells.

Fig. 68. Young spermatocyst from near distal end of follicle. Several connective-tissue cells surround the spermatogonia.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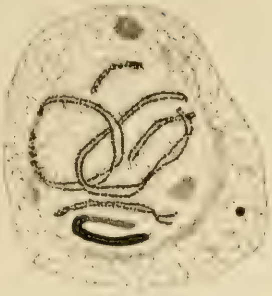
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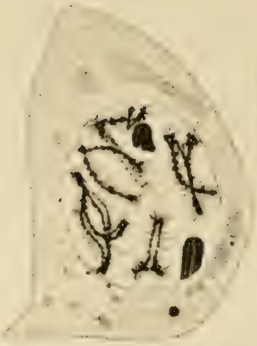
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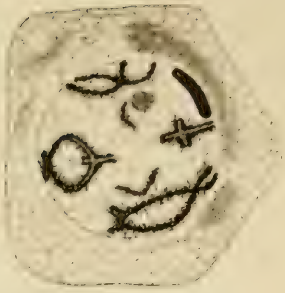
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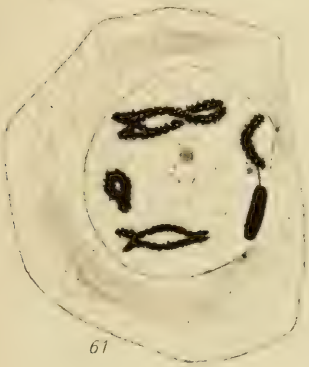
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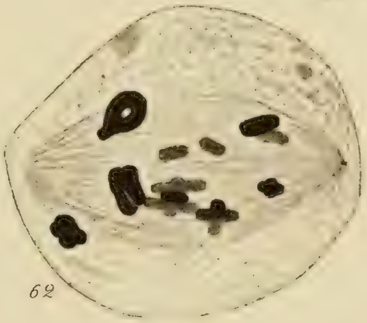
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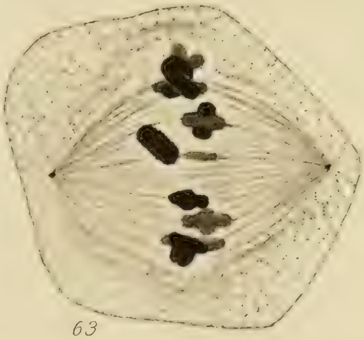
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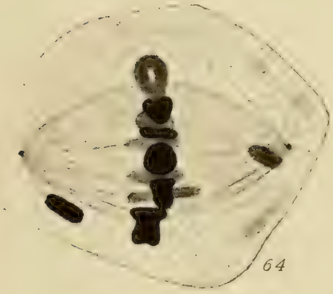
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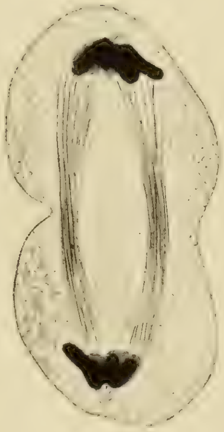
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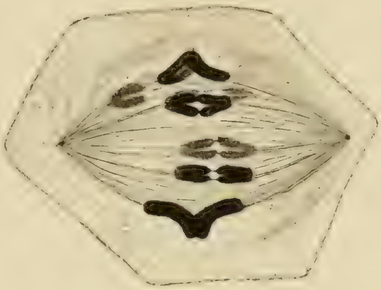
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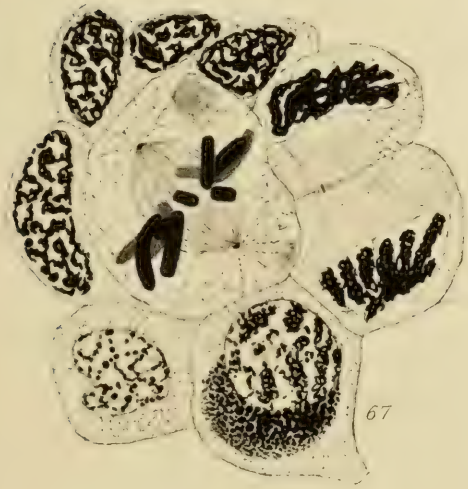
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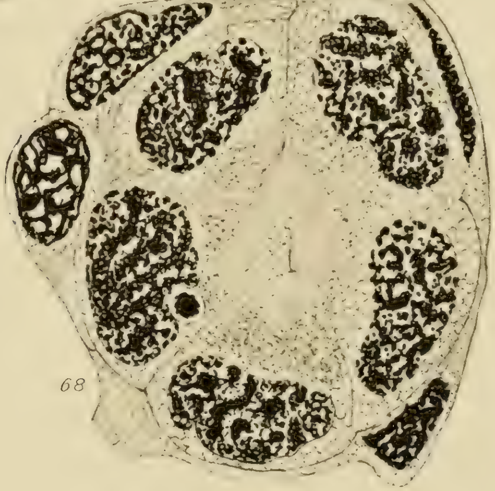
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H S D del.

PLATE 5.

Figs. 69–73. *Steiroxys trilineata.*

Fig. 69. Stage *e,* primary spermatocyte. The monosome is seen above and at the left.

Fig. 70. Stage *g,* primary spermatocyte. The monosome is seen at the lower side of the nucleus.

Figs. 71, 72. Stage *h,* primary spermatocyte. The monosome is distinguished from the autosomes by its more compact structure.

Fig. 73. Metaphase of first maturation division. The monosome is connected with only one pole.

Fig. 74. Metaphase, first division of *Melanoplus femoratus.* The monosome is seen at the right connected with only one pole.

Figs. 75–81. *Dissosteira carolina.*

Fig. 75. Semiresting stage of second spermatocyte. The monosome is seen below in the figure and at a lower focus.

Fig. 76. Prophase of second division.

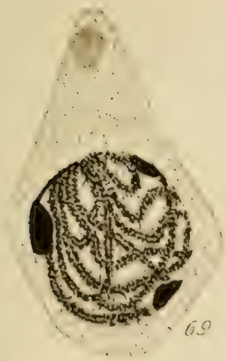
Fig. 77. Metaphase of second division.

Figs. 78–79. Anaphase of second division. The figures are from two successive sections of the same cell. All the chromosomes are shown.

Fig. 80. Telophase of second division.

Fig. 81. Spermatids. The monosomes still retain their compact structure.

Fig. 82. Spermatid of *Stenobothrus curtipennis.* The monosome is still compact.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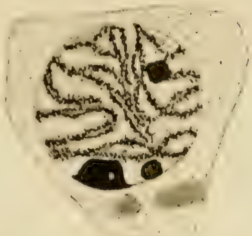
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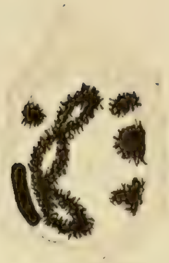
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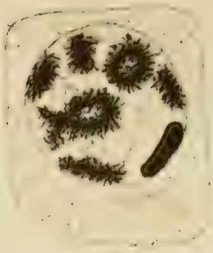
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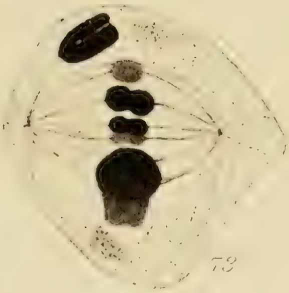
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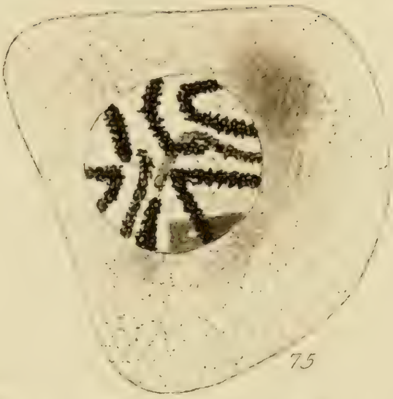
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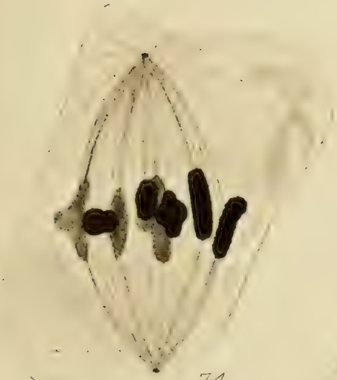
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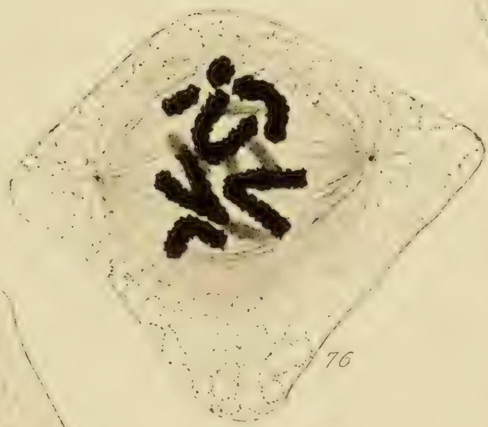
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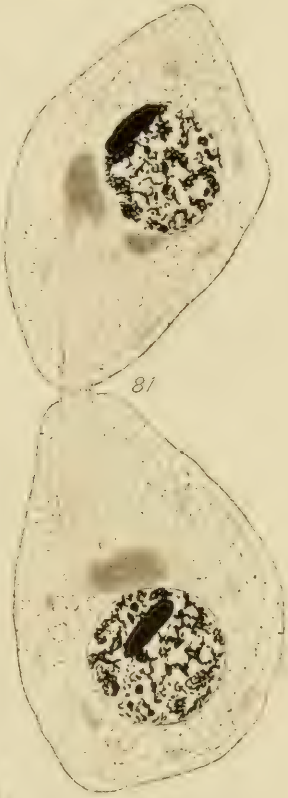
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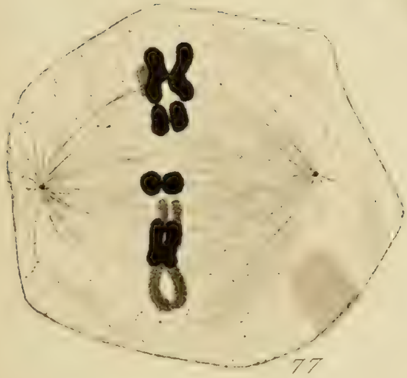
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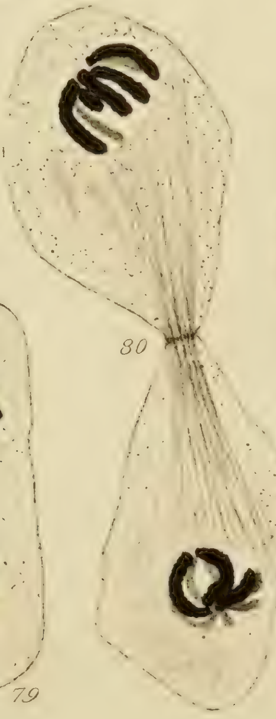
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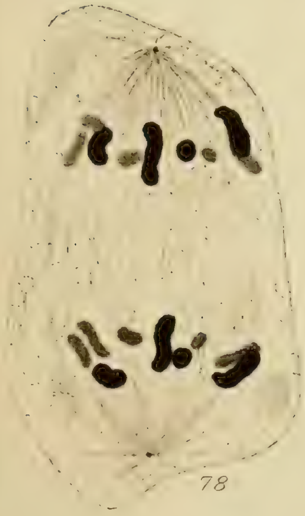
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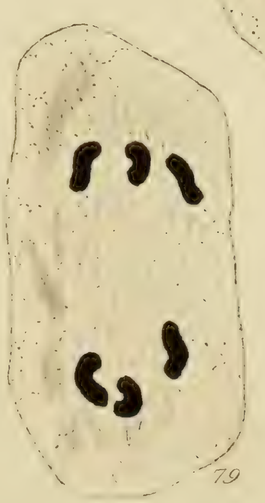
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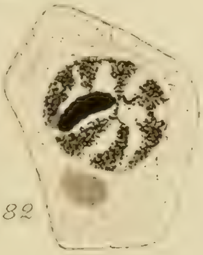
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H S D del.

PLATE 6.

All figures are of *Stenobothrus curtipennis.*

Fig. 83. Stage *g*, primary spermatocyte.

Figs. 84, 85. Stage *h*, primary spermatocyte. The monosome is seen below in Figure 85.

Figs. 86, 87. Stage *i*, primary spermatocyte.

Figs. 88, 89. Metaphase first maturation division. The figures are drawn from successive sections of the same cell, all the chromosomes being shown. The monosome is near the lower pole in Figure 88.

Figs. 90, 91. Metaphase of first division. The figures are drawn from successive sections of the same cell. The monosome is near the upper pole in Figure 91.

Fig. 92. Metaphase of first division. The monosome is near the lower pole.

Fig. 93. Anaphase of first division.

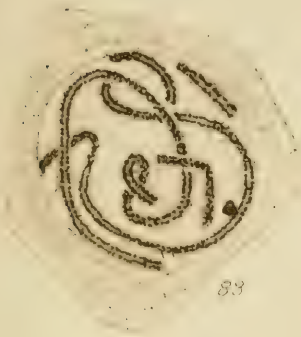
Fig. 94. Late anaphase of first division.

Fig. 95. Semiresting stage, secondary spermatocyte. The monosome is seen at the upper side of the nucleus.

Fig. 96. Metaphase of second division.

Fig. 97. Anaphase of second division.

Fig. 98. Telophase of second division.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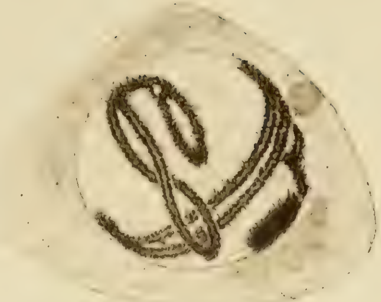
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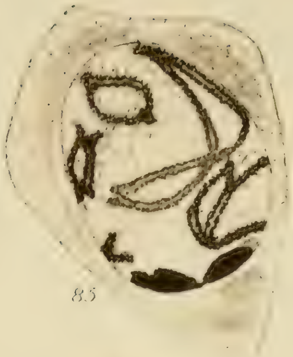
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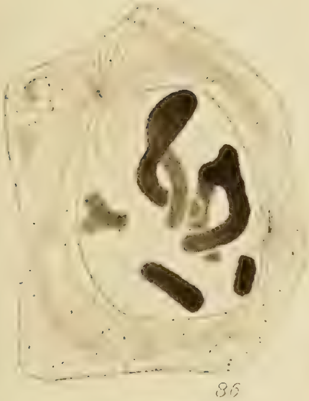
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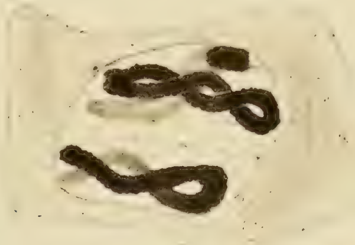
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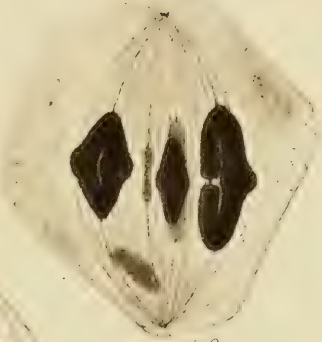
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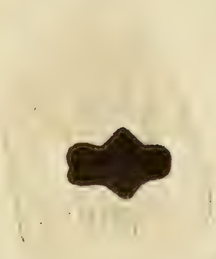
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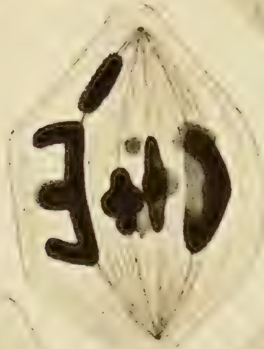
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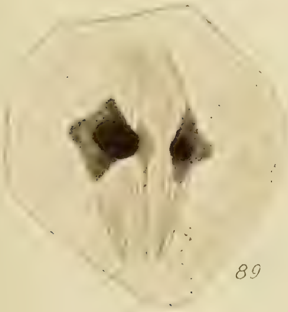
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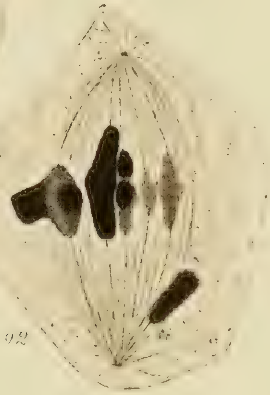
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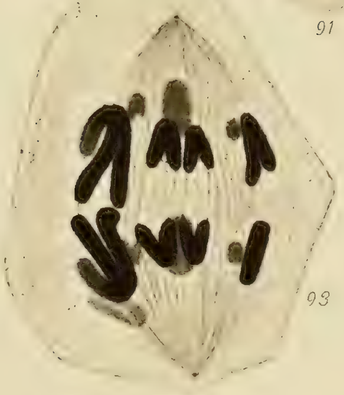
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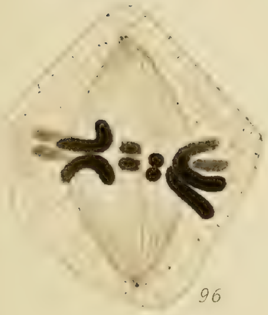
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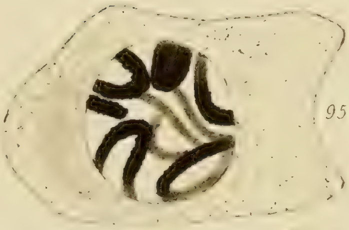
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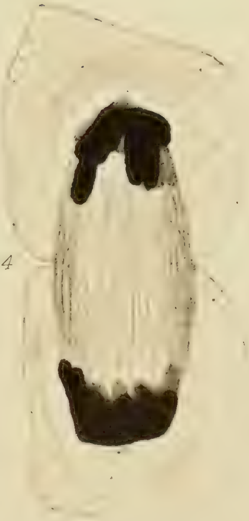
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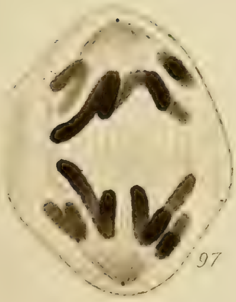
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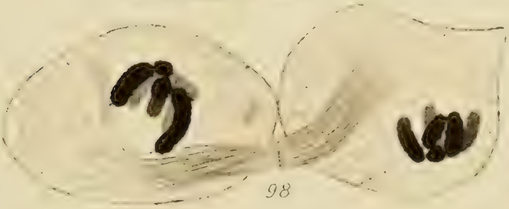
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PLATE 7.

Figs. 99–111. Successive stages of the monosome in the primary spermatocytes of *Dissosteira carolina.*

Figs. 112–114. Monosomes of *Arphia tenebrosa* during the late growth period.

Figs. 115–124. Successive stages of the monosome in the primary spermatocytes of *Melanoplus femoratus.*

Figs. 125–150. Successive stages of the monosome in the primary spermatocytes of *Stenobothrus curtipennis.*

Figs. 151–158. Successive stages of the monosome in the primary spermatocytes of *Steiroxys trilineata.*

Fig. 159. Rod-shaped tetrad of *Hippiscus tuberculatus.*

Figs. 160–182 *Dissostiera carolina.*

Figs. 160–164. Successive stages of the rod-shaped type of tetrads.

Figs. 165–168. Successive stages of the cross-shaped type of tetrads.

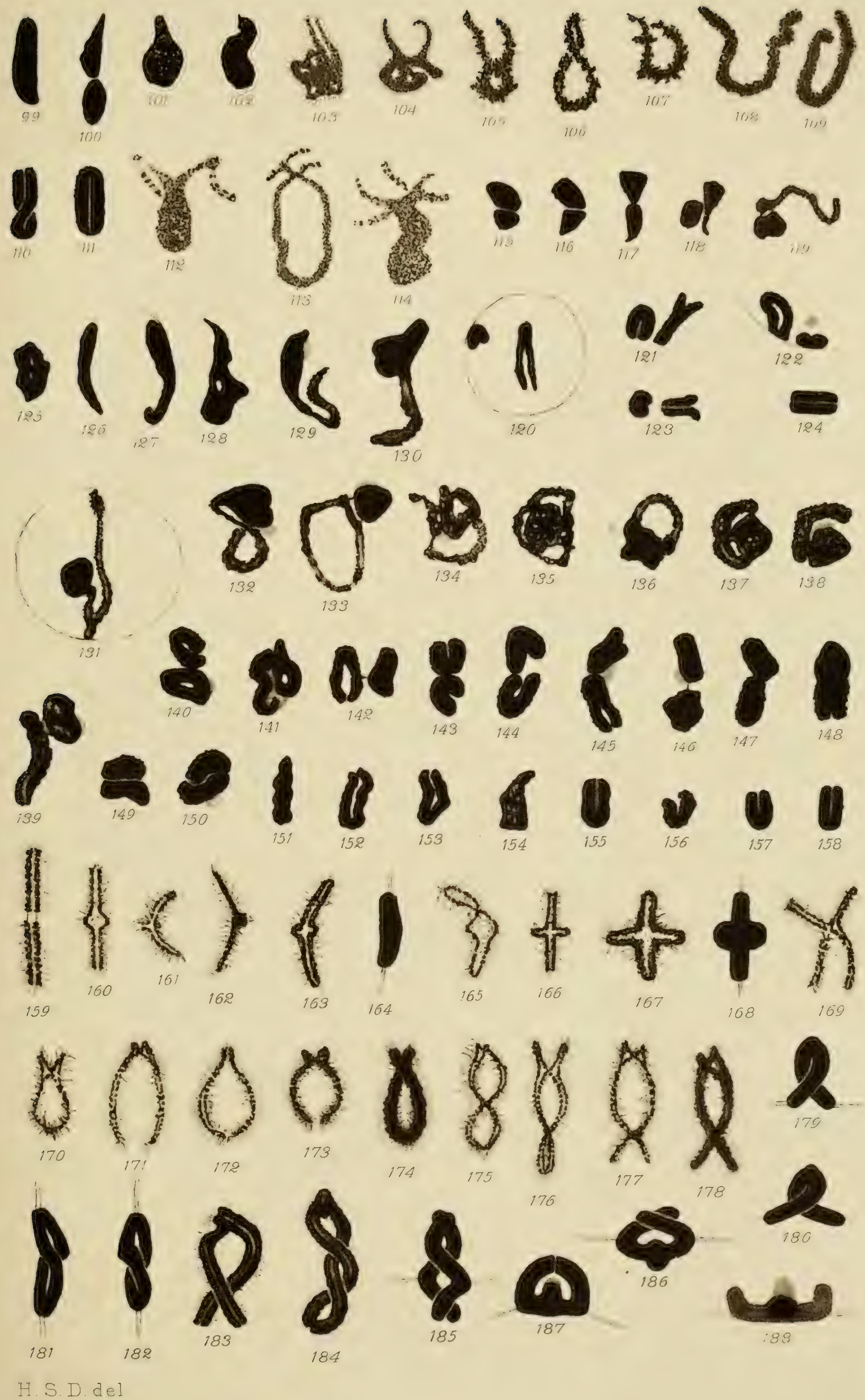
Fig. 169. Extreme development of cross-shaped type of tetrad.

Figs. 170–174. Successive stages of ring-shaped tetrads.

Figs. 175–182. Successive stages of the crossed loops.

Figs. 183–188. Large loops of *Stenobothrus curtipennis.*

PLATE 7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H. S. D. del

PLATE 8.

All figures are of *Steiroxys trilineata.*

Fig. 189. Polar view, metaphase of first maturation division. The rod-shaped chromosome is the monosome.

Fig. 190. Anaphase of first division. The monosome can be seen near the right pole.

Fig. 191. Early telophase of first division. The longitudinally split monosome has lagged behind the autosomes.

Fig. 192. Late telophase of first division.

Fig. 193. Semiresting stage of secondary spermatocytes. In one cell the monosome lies within, in the other without the nucleus.

Fig. 194. Prophase of second division. The monosome is seen below.

Fig. 195. Metaphase of second division.

Fig. 196. Early anaphase of second division in a cell which lacks the monosome.

Fig. 197. Later anaphase. The monosome is just dividing.

Fig. 198. Late telophase of second division. The monosome is not present.

Fig. 199. Late telophase of second division. The monosome is seen projecting from each mass of daughter chromosomes.

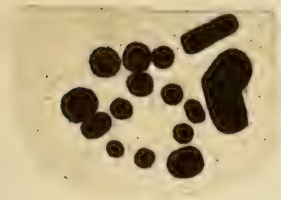
Fig. 200. Early spermatid. The monosome is still compact.

Fig. 201. Later spermatid. The monosome is lacking.

Fig. 202. Same stage as Figure 201, but the monosome is present.

Fig. 203. Same stage as Figure 201.

Figs. 204–212. Successive stages in the metamorphosis of the spermatid.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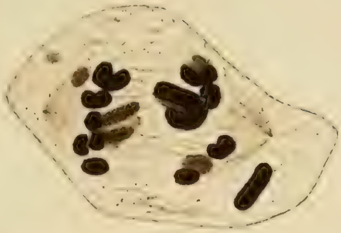
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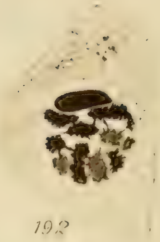
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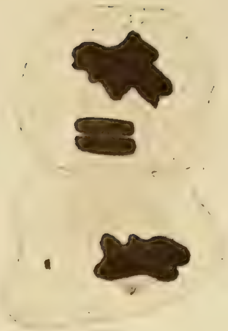
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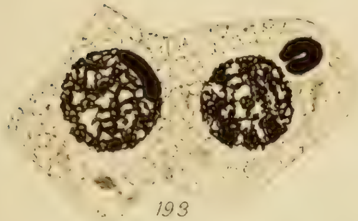
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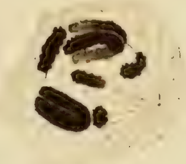
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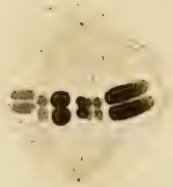
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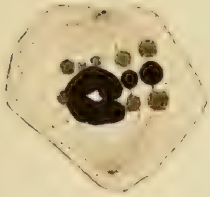
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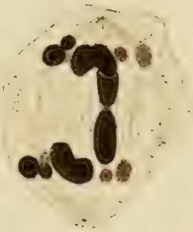
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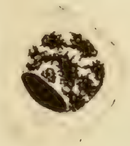
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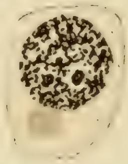
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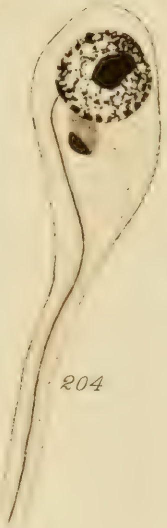
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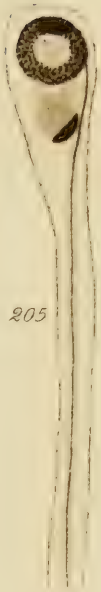
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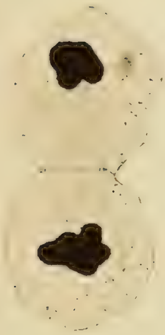
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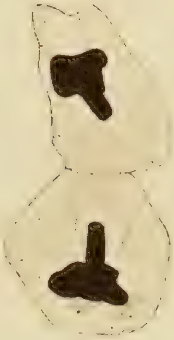
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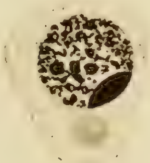
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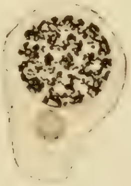
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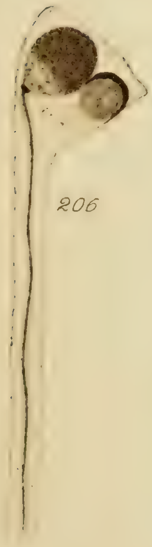
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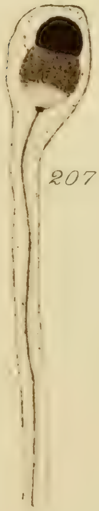
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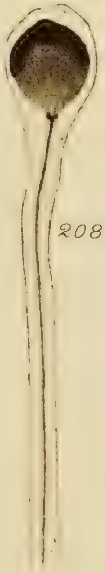
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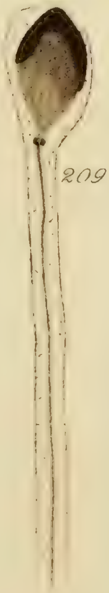
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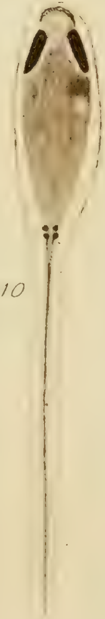
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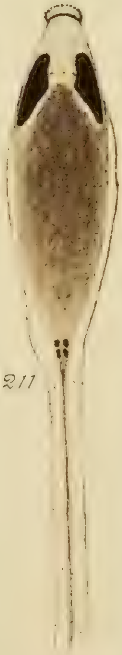
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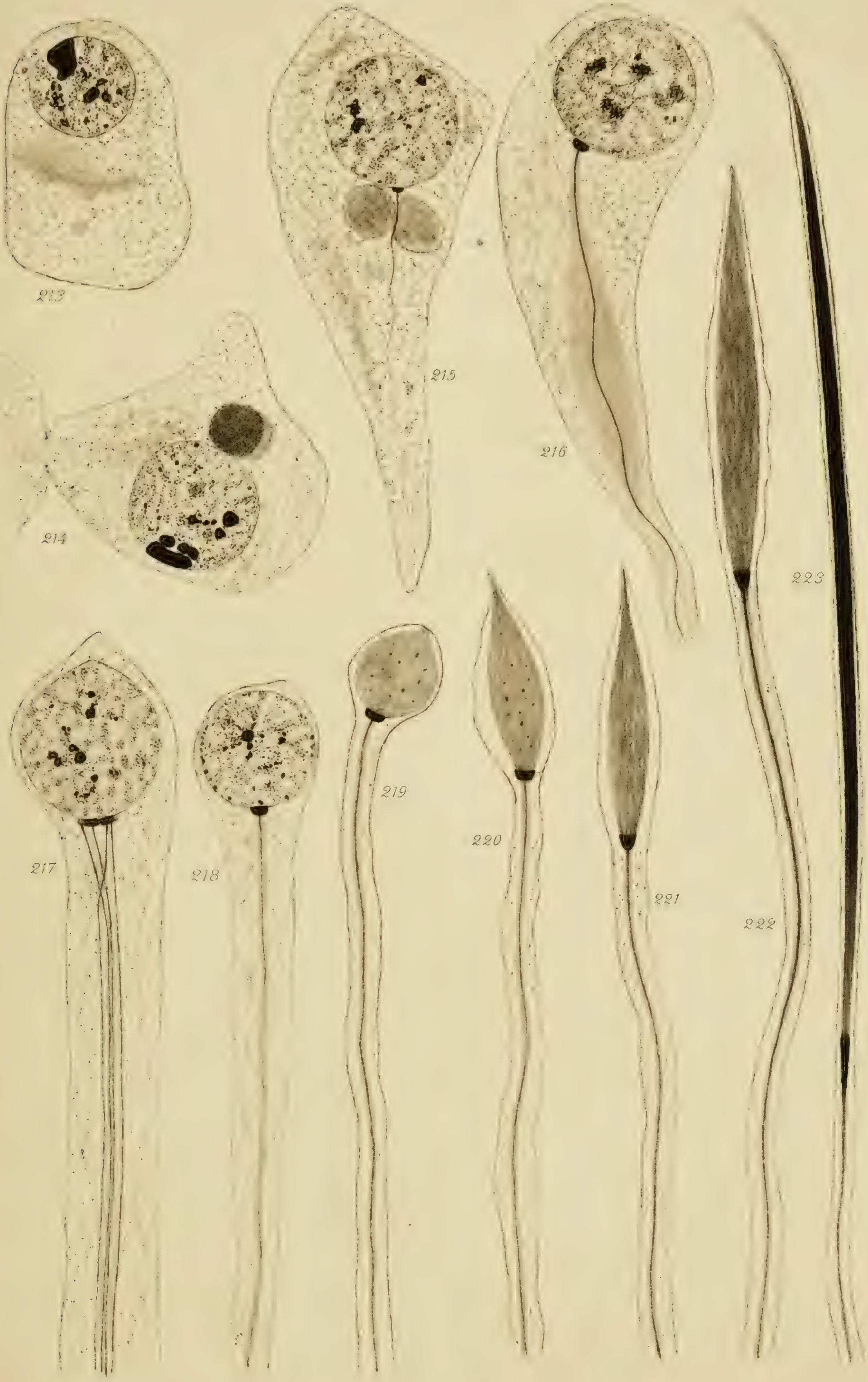
H. S. D. del

PLATE 9.

Figs. 213–223. Successive stages in the metamorphosis of the spermatids of *Dissosteira carolina*. For detailed account of Figures see text (pp. 110–113).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Bulletin of the Museum of Comparative Zoölogy
AT HARVARD COLLEGE.
Vol. LIII. No. 3.

MATURATION, FERTILIZATION, AND SEGMENTATION OF PENNARIA TIARELLA (AYRES) AND OF TUBULARIA CROCEA (AG.).

By George T. Hargitt

With Nine Plates.

CAMBRIDGE, MASS., U. S. A.:
PRINTED FOR THE MUSEUM
October, 1909.

No. 3.— CONTRIBUTIONS FROM THE ZOÖLOGICAL LABORATORY OF THE MUSEUM OF COMPARATIVE ZOÖLOGY AT HARVARD COLLEGE, UNDER THE DIRECTION OF E. L. MARK, No. 202.

Maturation, Fertilization, and Segmentation of Pennaria tiarella (Ayres) and of Tubularia crocea (Ag.).

BY GEORGE T. HARGITT.

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I. Introduction.

The literature on the embryology of the Coelenterata, and more particularly on that of Hydromedusae, contains very few articles dealing with cytologic details of early stages, though there are a considerable number dealing with the more general questions of cell division, later development, etc. Cytologists seem largely to have neglected these forms, which may be of considerable importance in a thorough understanding of cell structure and development. The few papers which deal with the early history of the egg of Hydromedusae show numerous gaps, and also suggest many points of difference between the Hydromedusae and other groups of invertebrates. The accounts of the history of the germinative vesicle during the periods of maturation and fertilization have been especially unsatisfactory and conflicting. It has therefore seemed worth while to study again with care these stages in the same forms as those upon which some of the earlier work was done.

The problem was suggested to me by my father, C. W. Hargitt, several years ago, and the work was started during 1906 at Syracuse University. The summers of 1907 and 1908 were spent upon the subject at the Laboratory of the United States Fisheries Bureau at Woods Hole, and during the past two years the work has been carried to completion in the Zoölogical Laboratories of Harvard University.

To Dr. F. B. Sumner, Director of the U. S. Fisheries Laboratory at Woods Hole, I am indebted for facilities in collecting material, and for laboratory privileges. To Dr. C. W. Hargitt I owe much in suggestion and enthusiastic interest, especially during the early part of the work; and to Dr. E. L. Mark I am indebted for constant and helpful criticism and supervision during the past two years. To all of these it is a pleasure to express my appreciation of their aid and interest. I have also had the privilege of examining the preparations and drawings of the early stages of development of the eggs of Hydractinia and Pennaria which will form the basis of a forthcoming paper by C. W. Hargitt and W. M. Smallwood, to whom my thanks are due for this opportunity.

II. Material and Methods.

Tubularia crocea was collected during November and June from a floating dock and adjacent piles near the mouth of the Charles River in Boston, and also in April and in June from the piles of the U. S. Fisheries dock at Woods Hole. Pennaria tiarella was obtained during July and August at Woods Hole. Colonies of the latter were collected and taken to the laboratory late in the afternoon, male and female colonies being kept in separate dishes. The medusae became free about seven o'clock in the evening. When the eggs had been discharged in sufficient numbers, spermatozoa were introduced by adding a pipetteful of the water from the dish containing the male colonies, in which the spermatozoa were very abundant and active. No trouble was experienced in getting the eggs fertilized, the desired stages being therefore easily obtained. In order to secure stages earlier than fertilization, it was necessary to remove medusae from the colony before the time of liberation. The medasae thus artificially set free were at once fixed, some of them 12–15 hours before liberation, others 10 hours before, and still others only a short time before that event.

For killing, the following fluids were used: Flemming's stronger mixture; aqueous solution of corrosive sublimate and acetic acid; Bouin's picro-formol; Zenker's fluid. In preserving Tubularia the corrosive-acetic mixture was used mostly, and gave excellent results. Staining after its use was brilliant. Other fixing methods were used for comparison with the corrosive mixture. For Pennaria, Flemming's, Zenker's, and Bouin's fluids all gave good results, though the last was the most satisfactory.

Heidenhain's iron hematoxylin, either alone, or followed by Congo red, orange G, etc., produced fine staining, and was used almost exclusively in working out the form and condition of nuclear constituents. Conklin's picro-hematoxylin did good service in staining achromatic structures, such as asters, centrosomes, and the like, and gave good results for the study of the nucleolus. Delafield's or Ehrlich's hematoxylin followed by eosin proved most satisfactory for the study of nucleoli. For purposes of comparison, several staining methods were used in every case, viz: hematoxylin and cochineal, Mallory's phosphotungstic acid hematoxylin, iron Brazilin, both Auerbach's and the Ehrlich-Biondi mixtures of acid fuchsin and methyl green. These last two stains were of some value when used for comparison with others, but I did not find that they are the safe indicators of chromatin that some authors have claimed them to be. For instance, one slide stained by this method showed the following distribution of red and green: in young oöcytes every part—cytoplasm, nucleus and nucleolus—green; in larger oöcytes, cytoplasm green, reticulum of nucleus brownish or maroon, nucleolus blue; in cells of the polyp, nucleus and nucleolus green, cytoplasm red; in the cells of embryos, chromosomes in a mitotic figure green, spindle fibres and asters red, cytoplasm maroon. Sometimes the cytoplasm and nucleus of an embryo cell were of nearly identical colors, while in other cells of the same embryo they were of different colors. Hence in some cases the red and green stains would give the same results, in regard to chromatin and non-chromatin, as those produced by other stains; while in other cases, the relation of the red and green colors would lead to results different, and indeed opposite, from those obtained by other stains. It is almost certain, I think, that the green in these mixtures is not a specific chromatin stain, but is modified in its action by several factors, probably physical as well as chemical ones, as has been maintained by Heine ('95–6), Mathews ('98), Fischer ('99), and Tellyesniczky (:05).

III. Observations.

A. Pennaria tiarella.

1. *Growing eggs.*—In the present investigation no attempt has been made to work out the oögenesis, except to a slight extent in the matter of the relation of the chromatin to the nucleolus. Smallwood ('99) and C. W. Hargitt (:00, :04^c) have paid some attention to oögenesis and growth phenomena; what I have seen is, in the main, confirmatory of their results. In addition to the absorption of oöcytes by the growing egg, I may mention another possible method of nourishment not considered by my predecessors. In the later stages of growth, when nearly all the oöcytes have been absorbed, the large eggs seemed generally to have several short pseudopodia attached to the entoderm of the manubrium. This suggests that during this period of growth nourishment may be obtained directly by absorption from the manubrium, much as in the case of Eudendrium (C. W. Hargitt, :04^b, Congdon, :06) and Clava (Harm, :03, Hargitt, :06).

In oöcytes before the growth period the chromatin of the nucleus is rather deeply stainable, and is arranged in masses near the periphery of the nucleus. These chromatin masses occupy a delicate linin network, which is connected to the nucleolus by fine strands. The nucleolus itself seems to be composed entirely of non-chromatic matter, for in Conklin's picro-hematoxylin mixture it stains yellowish, while the chromatin is bluish; in hematoxylin and eosin the nucleolus is red, the nuclear reticulum blue or purple. In the nucleolus at this stage, there are present one or more vacuoles, which may disappear during the growth period. In a few instances the nucleolus has taken some of the chromatin stain, appearing brownish in picro-hematoxylin, or slightly purplish in hematoxylin and eosin. It may therefore sometimes contain a little chromatin, though apparently this is not usually the case, provided the staining reactions are to be regarded as at all specific. In early stages of the oöcytes, the nucleus is central, but during growth it moves to the periphery, as shown by Smallwood ('99) and Hargitt (:00, :04^c).

The germinative vesicle in the growing egg shows a very faintly stainable reticulum, and the chromatin is apparently finely divided and diffused along the network. This condition, as is well known, exists in many Hydromedusae and in some other animals. The nucleolus at this stage is nearly spherical, usually excentric, often markedly so (Figs. 1–4), and may contain vacuoles, though more commonly it appears homogeneous. The nucleolus selects the plasma stain at this stage also. At the end of the growth period, the nuclear reticulum shows so little affinity for basic stains that there appears to be, so far as this test shows, no chromatin present in the entire nucleus. I can suggest no explanation for this peculiar condition of the chromatin at this period, but it is normal and characteristic of this stage.

The staining reactions of the nucleolus above described make it probable that this body is composed mostly, if not entirely, of non-chromatic matter. Since I have not studied the very early stages in the oögenesis, and do not know how the nucleolus originates, it is not possible to say that there is no direct relation between the nucleolus and the chromosomes. The fact that the nucleolus does not change its staining reaction during the growth period of the egg, while the chromatin becomes diffused and loses its staining capacity, means either that the chromatin does not enter the nucleolus, or, at least, that it does not enter as chromatin. The further fact that the nucleolus does not enlarge during the growth of the egg, makes it nearly certain that the chromatin does not then enter it. It is likewise true that the concentration of the chromatin of the germinative vesicle into granules and strands just before polar cells are formed, is not necessarily coincident with the disappearance of the nucleolus (Figs. 1, 3). In some cases the two processes take place at different times, so that it seems probable that chromatin and nucleolus have no fixed relation to each other.

The time of disappearance of the nucleolus is not constant. For example, Figures 1 and 2 are from eggs of the same stage, having been killed ten hours before the usual time of liberation of the medusa; yet in one case the nucleolus is large, apparently having undergone little diminution of size, and in the other it has almost disappeared. Figures 3 and 4 also represent eggs of the same age; here the differences in the size of the nucleoli are still more marked. A comparison of these two sets of eggs also makes clear the lack of agreement between the disappearance of nucleoli and the appearance of chromatin strands. While the eggs are apparently of similar age, the chromatin appears to be more concentrated in those cases (Figs. 1, 3) where the nucleolus is larger, more diffuse where the nucleolus is smaller (Figs. 2, 4). I have no stages intermediate between these four and those in which the nucleolus has already disappeared. Figures 5–7 show that when the germinative vesicle has advanced further toward the formation of polar cells no nucleoli are present. It seems clear from Figures 1–7 that the nucleolus disappears gradually, perhaps by dissolving rather than by breaking up into fragments.

With regard to the nucleolus, then, the evidence seems clearly to lead to these conclusions: (1) it is mainly, if not entirely, a non-chromatic body at all stages from the young oöcyte to the mature egg; (2) it entirely disappears, probably by dissolving, before the nuclear membrane has been ruptured; (3) the time of its disappearance varies in different eggs; (4) there is a direct connection between the nucleolus and the linin network, so that an exchange of material may take place between nucleolus and chromatin along the linin network, but not in the form of prepared chromatin.

2. *Formation of Polar Cells.* Before describing the details of polar-cell formation something should be said regarding the time of its occurrence. In examining living eggs just after they had been discharged from the medusa (this was done by compressing the eggs slightly under a cover glass), the nuclei appeared small and without nucleoli. Evidently the polar cells had already been formed, though they were no longer discoverable, doubtless because they had been lost in the water. Eggs similarly examined a few hours before the discharge of the medusae show the germinative vesicle, which usually contains a nucleolus. The conclusions reached from the study of fresh eggs were confirmed by the examination of sections of fixed eggs.

Those which were killed a few hours before their discharge possess the germinative vesicle, and those killed after their discharge show the small egg nucleus. Since eggs killed immediately before their discharge from the medusa are the only ones that have shown polar cells in process of formation, it may be confidently stated that polar cells are usually formed only during this brief period, as Hargitt (**:04**^{c}) has already suggested. While all the maturing eggs of one medusa are, as a rule, in approximately the same condition, this is not invariably the case, for while some are actually forming polar cells, others still have large germinative vesicles. This I take to be evidence of individual differences in the eggs themselves.

In the eggs of Pennaria at the end of the growth period the germinative vesicle is close to the outer surface of the egg, being covered by only a very thin layer of cytoplasm; the chromatin, in the form of fine grains, is distributed along the reticulum, and shows only a slight affinity for the usual nuclear stains. The nucleolus behaves in the way already described, disappearing before the membrane of the germinative vesicle does. The time at which the chromatin begins to concentrate, preparatory to forming chromosomes, is variable. Figure 1 represents an egg killed ten hours before the time when presumably it would have been discharged, and shows the chromatin more concentrated and more deeply stained in one part of the germinative vesicle than in other parts. Figure 2 represents the germinative vesicle of an egg which is only slightly more advanced than that of Figure 1. The chromatin is diffused, there being no sign anywhere of deeply staining granules. The shape of the vesicle in this case is one often assumed just before the formation of the maturation spindle. Figures 3 and 4 show the condition of the germinative vesicles of two eggs killed just after the liberation of the medusa; the concentration of chromatin into granules along the reticulum is well exhibited. The chromatin in Figure 4 is only slightly more concentrated than that in Figure 1, although the latter was killed ten hours earlier. The same difference in the time of concentration of the chromatin is shown in Figures 3 and 7; the former, representing the germinative vesicle, has the chromatin much more concentrated than the latter, which represents a later stage, in which the maturation spindle is forming.

Figures 3 and 4 show a wrinkling of the membrane, which seems to be the result of a decrease in size of the germinative vesicle. The concentration of chromatin is also beginning, and these eggs are undoubtedly near to the time of polar-cell formation. The Figures also make it plain that the size and staining capacity of the nucleolus is independent of the concentration of the chromatin. No centrosomes or radiations have yet appeared in connection with these germinative vesicles. Before the chromosomes begin to form, the germinative vesicle becomes smaller, and often ellipsoidal, and the cytoplasm forms a special zone about it (Figs. 5, 6). The conditions shown here, especially the presence of the cytoplasmic envelope, are those which immediately precede the formation of the maturation spindle, for they occur during the following stages (Fig. 7) when the spindle is forming, but are not present in the earlier germinative vesicle (Figs. 3, 4). It should be said that the ellipsoidal shape of the germinative vesicle may be found some time before the spindle appears, eggs killed ten hours before the time of discharge sometimes having this form (Fig. 2). Figures 5 and 6 show only a slight concentration of the chromatin, and neither centrosomes nor radiations. The next older stage found (Fig. 7) already possessed centrosomes with well marked astral rays in the cytoplasm, and the maturation spindle was beginning to be formed. The cytoplasmic envelope surrounding the germinative vesicle was less evident than in the preceding stage. The membrane of the vesicle was deeply indented opposite one aster, and slightly so opposite the other. This may be taken as evidence that the maturation spindle is formed, in part at least, from the cytoplasm, and that it may aid in the dissolution of the nuclear membrane.

The next stage which I have found shows the first maturation spindle with the chromosomes at its equator. In this stage the spindle is approximately tangential to the nearest point of the surface of the egg, though one pole is sometimes a little closer to the surface of the egg than the other. The polar radiations are less distinct than formerly, though the centrosomes are still present. Both centrosomes and radiations were at first overlooked, and in a previously published note (G. T. Hargitt, :09) they were stated to be absent. Figures 8a, 8b, 9a, 9b, 10a and 10b are nearly polar views of the equatorial-plate region of three eggs, the chromosomes in each case extending over two sections. Figure 11 (Plate 2) is a similar view of a fourth egg, though here the chromosomes occupied three sections. In Figures 9 and 10 several of the chromosomes are seen to be in the tetrad condition, and a splitting of some of them seems to have begun. Others appear as short rods, but are so small that the direction in which the division occurs cannot be accurately determined. Figure 11, however, makes this somewhat clearer. Here are found bodies in rod-, x- and v-shaped conditions, as well as some which have a roundish or irregular form. The impression made by the appearance of the chromosomes is that the rods split longitudinally, beginning at one end, though the evidence is not sufficient to make this certain. After the division has been completed the parts appear to arrange themselves in the form of an x. The final disposition of these bodies or their parts was not determined, since the stages immediately following are lacking in my preparations.

An accurate count of the chromosomes was not possible, because, unfortunately, in every case the chromosomes occupied two or three sections. By careful focussing to determine which had possibly been divided in sectioning and were therefore to be found in two sections, and by making accurate camera drawings of all the chromosomes of the two sections possibly involved, and superposing them, it has been possible to make a reasonably safe estimate of the number of separate elements. In one egg there were clearly ten. Another case, while not so satisfactory, makes it safe to say that not more than ten are present. By a similar method of estimate a third case gave eleven, and a fourth thirteen, as the probable number of separate chromosomes. A comparison of the Figures shows clearly that some of the chromosomes are beginning to split, so that probably what appear to be separate elements are really moities of a single body. Just how many are split sufficiently to give this appearance I could not determine. In Figure 11 the splitting has proceeded so far, and the chromosomes are so closely massed, that no safe estimate of the number of chromosomes could be based on this egg. On the whole, the evidence seems to point to ten as the probable number of chromosomes in the spindle. This is a smaller number than occurs in the cleavage spindle, so that there is probably a reduction in number before this stage is reached. The accurate determination of the number of chromosomes in the second cleavage spindle is, however, more difficult than in the maturation spindle, so that I cannot state with any degree of confidence the number of chromosomes which occur in the former. The reduction had already occurred before the first maturation spindle was formed, but as to where and when, I have no direct evidence.

The spindle, at an earlier period nearly tangential, now assumes a radial position. The asters, which were smaller during the tangential position than when first formed, entirely disappear from both poles. The centrosomes, however, remain, and the spindle then appears as shown in Figure 12. The chromosomes here are somewhat scattered, and an accurate count is not possible. Near the more superficially located pole, the chromosomes seem to have become massed together and to have left their normal place, being at one side of the spindle, with which, however, they are still connected by fine filaments. The polar cell would soon have been formed in this egg. After the first polar cell is detached, the chromosomes at the deep end of the spindle seem to lose their identity, and form a more or less typical resting nucleus, a condition which will be described more at length a little later (p. 171).

The actual formation of the second maturation spindle I have not observed, the stage represented in Figure 13 being the earliest condition subsequent to that of Figure 12 which I have found. The first polar cell (Fig. 13) is shown, still connected to the egg by interzonal filaments; the chromosomes have already lost their individuality and their substance is loosely distributed through the outer end of the polar cell, though not constituting a definitely formed nucleus. The second maturation seems to represent an advanced stage of the spindle. In the outer half the fibres are more prominent than those within the egg. The latter converge more or less sharply toward a centre within the egg, while the outer fibres converge less definitely toward the distal end of a somewhat conical protuberance of the surface of the egg, which one naturally interprets as the prospective second polar cell. Within the egg faint astral radiations appear in the region of the apparent pole of the spindle, but no such radiations are found at the end of the peripheral fibres. At neither pole is there a body resembling a centrosome. As regards the chromatin of the second polar cell, the Figure suggests two possibilities. Either (1) the chromatin may be distributed along the fibres of the peripheral half of the spindle, in which case these fibres represent true spindle fibres with some chromatin, and the inner half of the fibres represent interzonal filaments. This would, perhaps, account for the prominence of the fibres in the polar cell, and would not be greatly unlike the condition in the adjacent first polar cell. Or (2) the chromatin that normally belongs to the polar cell may be represented in the two large chromatic masses near the equator of the spindle, which have not been drawn into the polar cell, and apparently would have been left in the egg had not the constriction, already started, been interrupted by the killing of the egg. This latter view gains some weight, perhaps, from the aberrant condition of part of the peripheral group of chromosomes seen at a slightly earlier stage in the first maturation spindle (Fig. 12). It may be that part of the chromatin of the polar cell is in the two chromatin masses, and that another part of it is distributed along the peripheral fibres. Whatever the view regarding the chromatin, it seems fairly certain that the thickenings in the equator of the spindle represent the interzonal bodies of ordinary cell division, and the spindle is therefore in an advanced stage. The number of chromosomes in this egg cannot be stated with confidence, though the six spherical bodies are certainly egg chromosomes, and several smaller, deeply staining bodies near by may be of similar import. The number of chromosomes left in the egg after the formation of the second polar cell cannot be more than ten, and may possibly be less.

While these maturation figures leave much to be desired in clearness and ease of interpretation, they do show without doubt that the polar cells are formed by a more or less regular mitosis.

A somewhat unusual condition is shown in Figure 14. Two other eggs in the same medusa have large ellipsoidal germinative vesicles without nucleoli and with a dense cytoplasmic layer about the germinative vesicle, such as is shown in Figures 5–7, which represent the condition occurring just before the formation of the first polar cell. A fourth egg in the same medusa has the nucleus in the condition shown in Figure 18. The eggs in this medusa were consequently at a stage corresponding very nearly with that of the formation of the polar cells. The nucleus shown in Figure 14 is apparently in the so-called resting condition, the chromatin being in the form of a diffuse and feebly staining network. Strongly marked cytoplasmic radiations are directed to a point at the surface of the egg which is slightly peripheral to the outer end of the nucleus; but no centrosome could be found at this point, and no other radiations were present in the egg. Outside the egg, and also outside the jelly-like membrane which surrounds it while it is within the medusa, is a body which closely resembles the first polar cell shown in Figure 13. There is at one end a deeply staining granular mass, apparently chromatin, and stretching from this mass toward the opposite end of the body there is an arrangement of the granules which gives the body a faintly striate appearance. The fact that this body is outside the membrane rather than inside it, as in the polar cells shown in Figures 12 and 13, may be an argument against considering it a polar cell; but it seems not unreasonable to suppose that the body may have been forced through the jelly-like envelope, which in the living animal is probably soft. Considering all the evidence, it is hard to escape the conclusion that we have here a stage between the formation of the first and the second polar cells. If so, then the chromatic matter of the deep half of the first maturation spindle has been reorganized into a nucleus with a definite membrane, the chromatin having become diffused as in the typical resting nucleus. The radiations are perhaps the first indication of the second maturation spindle.

Another egg (Fig. 15) shows essentially similar conditions, and renders the above explanation more probable. In this case a small body is found on the surface of the egg, inside the envelope which surrounds the egg. A dark mass within this body probably represents chromatin, the entire body being a polar cell, which agrees in position with the polar cell shown in Figure 13, though somewhat smaller. The chromatin left in the egg after the formation of the first polar cell has been reorganized in this case into several vesicles, each with a definite membrane; a reticulum has been formed in each vesicle, with the chromatin more or less scattered, but not so diffusely as in Figure 14. Radiations, similar to those found at the peripheral end of the nuclear body in Figure 14, are present here also, though not so plainly marked. At the inner border of the group of more deeply lying vesicles are faint astral radiations. These radiations probably represent the asters at the poles of the forming second maturation spindle.

Since these are the only instances of such conditions which were found, it is not possible to say whether such a stage in the formation of the polar cells is typical. However, this lack of evidence is not a fatal objection to the interpretation given, since stages showing maturation spindles are almost as rare; indeed, only one egg showed a second polar cell in process of formation. Moreover, it is significant that conditions similar to these are quite common in Tubularia, though the polar cell was not found in these cases.

The earliest stages in the reorganization of the chromosomes into an egg nucleus, after the formation of the second polar cell, I have not succeeded in finding. Somewhat later the egg nucleus is represented by several vesicles (Plate **3**, Figs. 21, 22). Each of these contains chromatin in a network of linin, though often the chromatin is collected most abundantly at the nodes of the net or close against the nuclear membrane. Sometimes these vesicles soon fuse with one another, but they may remain separate and distinct till about the time of their union with the sperm nucleus.

To sum up: polar cells appear to form just before, or at the time of, liberation of the medusae. The concentration of chromatin in preparation for the production of chromosomes is not coincident with the disappearance of the nucleolus. The germinative vesicle becomes ellipsoidal, asters with centrosomes appear, and the maturation spindle forms, in part at least from the cytoplasm, before the membrane of the germinative vesicle disappears. The first maturation spindle is at first tangential to the surface of the egg, and contains the reduced number of chromosomes, probably ten. A splitting occurs, resulting in x- and v-shaped figures. The spindle becomes radial, the asters are lost, though the centrosomes remain, and the first polar cell is formed. There is some evidence that before the second maturation spindle forms, the chromatin remaining in the egg reorganizes into a nucleus with a definite membrane, the chromatin taking on the condition typical of the so-called resting stage. A second maturation spindle and a second polar cell are formed. The chromosomes remaining in the egg form the egg nucleus. This is usually composed of several distinct vesicles, which may fuse at once or remain distinct till the time of conjugation with the sperm nucleus.

3. *Fertilization.*— When spermatozoa are present in considerable numbers, they may entirely surround the living egg, but nearly always they appear to be more plentiful at one or two places than elsewhere. In some of the living eggs a very definite cone on the surface of the egg was seen at the place where the spermatozoa were most abundant, and in several cases there were apparently two cones present in one egg, spermatozoa being collected about both of them. Whether the cone is a true "cône d'attraction" or a "cône d'exsudation" (Fol, '79) could not be determined on account of the impossibility of ascertaining whether it was formed before or after the entrance of the spermatozoön. An entrance cavity, such as was found in coelenterates by Metschnikoff and by Brauer, I have never seen. After the medusae were liberated, the eggs sometimes retained their position on the manubrium. When, in such cases, spermatozoa were present in the water, some of them gained access to the ova, for sections of such eggs showed that fertilization had occurred under these circumstances. It is probable that the tissues surrounding the eggs had been ruptured before the spermatozoa gained access.

Hargitt (:00) speaks of a sort of "convulsive surface torsion" of the egg of Pennaria soon after the spermatozoön enters, and I, too, found that under the same conditions the surface of the egg became irregular, and then after a short time apparently rounded out again. Sections show that about the time of entrance of the spermatozoön the cytoplasm is extremely active; protuberances are extended from many points of the surface of the egg, some rounded, others elongated. Some of these seem to become entirely detached, whereas others are apparently withdrawn. Somewhat later, when conjugation of the germ nuclei is about to occur, the surface of the egg as a rule appears regular and smooth.

As has already been stated, the polar cells arise before the eggs are discharged, and hence usually before the spermatozoön enters. In a single case sections showed an apparent exception, the spermatozoön having entered an egg while it was still within the medusa, and before the polar cells had been detached. Figure 18 (Plate 2) represents the condition referred to; the smaller vesicle appears to be the sperm nucleus, the larger one has every appearance of a germinative vesicle. As arguments that the latter is such, the following points may be suggested: (1) the egg nucleus is usually not so close to the periphery of the egg; (2) the size and shape of the nucleus are characteristic of germinative vesicles just before the first maturation spindle forms (Figs. 5, 7); (3) at least one, and possibly two, astral centers are present (somewhat like Figure 7), a condition not usually found accompanying egg nuclei; (4) other eggs in the same medusa had their nuclei in a condition which precedes the formation of the maturation spindles.

Sections show that while the spermatozoön may enter the egg at any point, it more commonly enters rather close to the egg nucleus (Fig. 18). The cones seen at the surface of living eggs may also be recognized in those which have been fixed (Figs. 17a, 19b), though this is not always the case (Figs. 18, 19c). Sections of eggs which showed this cone also showed a spermatozoön or sperm nucleus, consequently the cone is to be considered as an entrance cone (Wilson, :00) rather than a true attraction cone.

Sometimes there is located in the periphery of the egg, and extending from its surface to the sperm nucleus, a differentiated, funnel-shaped region (Figs. 17a, 20a), which one naturally refers to the influence of the spermatozoön. Sometimes the axis of the funnel or cone-shaped region is straight and nearly radial (Fig. 17a), at other times it is more or less curved (Fig. 20a), but its apex coincides with the sperm nucleus, and its broad basal end is always at the periphery of the egg. Similar phenomena, apparently due to similar causes, have been observed in other eggs. They are especially prominent in the eggs of certain amphibians, where the pigment renders the condition more obvious (Schultze, '87, Fick, '93, King, :01), and have also been observed in the earth worm (Foot, '94), in echinoderms (Wilson, '95), etc. An angle has sometimes been found in this "track"; it was interpreted by Roux ('87) as the end of the "entrance path," after which the sperm comes under the influence of the egg nucleus and moves toward this in the "copulation path"; while Fick considered the angle due to the rotation of the spermatozoön. The "track" in Pennaria never showed this angle, and as a rule was nearly radial and short, leading directly to the egg nucleus, which was usually near the point of entrance of the sperm. In many cases no "track".

was seen, though this may not be remarkable, since the region is discernible only by a slight difference in the staining reaction. Since an aster seems not always to accompany the spermatozoön, it cannot be definitely determined whether there is a rotation of the sperm during its migration, though when present the aster is sometimes at the deeper side of the spermatozoön. Even when this is the case no angle is found in the "path" (Fig. 17a).

Usually the sperm head penetrates the egg only a short distance before it begins its transformation into the sperm nucleus, for one finds the latter in its early stages not far removed from the periphery of the egg (Figs. 18, 19b, 19c). This is characteristic and appears to occur in all cases except where supernumerary spermatozoa enter the egg, in which case the extra spermatozoa remain unchanged for some time. After the entrance of the spermatozoön there is often an entire absence of asters in connection with the germ nuclei; this makes it impossible to distinguish the egg nucleus from the sperm nucleus, or to learn exactly what transpires before they conjugate.

Figure 16a represents the sperm nucleus, and Figure 16b the egg nucleus of the same egg, both being near the surface of the egg. Between the surface and the sperm nucleus there can be traced in the sections an ill defined "entrance track." The nucleus itself has a definite aster, the rays of which centre in a large clear area, in which, however, no central body is found. This area is on the side of the nucleus opposite to that which faces the egg nucleus; however, a second very faint and indefinite series of radiations seems to exist on this side also. The sperm nucleus (Fig. 16a) is smaller than the egg nucleus (Fig. 16b) and spherical, while the latter is ellipsoidal and is not accompanied by any radiations. Each nucleus is composed of a single vesicle, and the chromatin is incorporated in a linin reticulum; it is more concentrated in the sperm nucleus. In this egg it is perfectly clear that the sperm nucleus possesses an aster and the egg nucleus none, though the aster is on the side opposite the egg nucleus, instead of the one facing it, as commonly occurs in the sperm nucleus of most animals. Perhaps a rotation of the sperm nucleus is yet to take place.

Figures 21a, 21b (Plate 3) represent the germ nuclei of another egg. These are close to each other, and not far from the surface of the egg. While one (Fig. 21a) is distinctly lobed, as though it had resulted from the fusion of a number of vesicles, the other (Fig. 21b) shows still more plainly that it has been produced by the confluence — not yet completed — of several separate vesicles. A fairly conspicuous and even-meshed network fills out each of the nuclei; but the chromatin is much more conspicuous in one of them (Fig. 21a) than in the other. A single well marked aster, without visible centrosome, sustains an interesting relation to the vesicles of the compound nucleus. It lies somewhat nearer the surface of the egg than the three more or less confluent vesicles, and the more pointed ends of these appear as though drawn out in the direction of the centre of the aster. This aster is on the side of the vesicles which faces the other nucleus, in connection with which there is no aster or other cytoplasmic differentiation. In the absence of an entrance cone or of a cytoplasmic "track," which, as we have seen, are characteristic of some sperm nuclei, it is difficult to decide which is the sperm nucleus. In another egg the aster was found to accompany the sperm nucleus (Fig. 16a), but, on the other hand, the multi-vesiculate condition is more characteristic of the egg nucleus. The compound nucleus, the one accompanied by an aster (Fig. 21b), has a shortest diameter of about 20 μ, whereas the other nucleus is only 9 μ in diameter; this seems to give evidence that the compound one is the egg nucleus. But even if this is so, the aster cannot be considered as belonging to the maturation spindle, since it is too prominent and not in the proper position, but must represent a new aster, which may perhaps persist to help form the cleavage spindle.

The most extreme condition of distinct vesicles representing the germ nuclei, though not the only such case found, is shown in Figure 22. This Figure is compiled from two sections by a careful superposing of camera drawings, and shows all the nuclear bodies found in the egg. Three of the vesicles (*a*) lay wholly in one section, and within 10 μ of the surface of the egg; the other vesicles (group *b*) were limited to the preceding section, and all were less than 20 μ from the surface. All the vesicles were in a zone of more deeply staining cytoplasm, which extended to the surface of the egg, and no astral radiations were present in connection with any of the vesicles. Spermatozoa were present on the surface of the egg, and one had probably penetrated, since no attraction cone was present, and the surface of the egg showed the irregularities which we have seen to be characteristic of the time immediately following the penetration of a spermatozoön. As neither sperm head nor other nuclear bodies were found in the egg, the vesicles figured must represent both sperm and egg nuclei. However, it is not possible to distinguish between the two.

Polyspermy occurs frequently in Pennaria, and in such cases each sperm head is accompanied by a distinct aster, though the aster is usually lost when one of the sperms begins to metamorphose into a nucleus. Perhaps the presence of the aster in polyspermic eggs gives additional reason for the conclusion that the aster, when present, owes its origin to the spermatozoön, though it seems as though the egg nucleus in some cases (Fig. 21b) possessed an aster, and the sperm nucleus none. Figure 17a represents a spermatozoön which has recently entered an egg. A cone is found at the surface of the egg, and there is a funnel-shaped region in the cytoplasm between the surface and the spermatozoön. In front of the sperm is an aster with no visible centrosome. Two other spermatozoa had penetrated the egg, but these remained in the most superficial layer of the cytoplasm, and showed no signs of migration. The egg itself had in each of these cases a cone, and near each spermatozoön a minute aster. The spermatozoön shown in Figure 17a was about 70 μ distant from the egg nucleus (Fig. 17b), while the two supernumerary spermatozoa were each over 100 μ distant from it. The relative nearness of the sperm figured, the large size of the aster, and especially the modified condition of the cytoplasm between the spermatozoön and the cone, make it appear probable that this is the one which would have been efficient in fertilization.

In polyspermic eggs it is not always the case that only one spermatozoön becomes a nucleus. In Figures 19a, 19b and 19c are shown three nuclei which occur in one egg. Figure 19a, the largest one, represents the egg nucleus with its chromatin evenly distributed in a reticulum, and with no evidence of concentration of the cytoplasm or of astral figures in its vicinity. Figures 19b and 19c are two sperm nuclei of about equal size, each of about one-half the diameter of the egg nucleus, and very close to the surface of the egg; the chromatin in each case being evenly distributed, as in the egg nucleus. As shown in Figure 19b, a cone marks the end of the egg radius in which lies one of the sperm nuclei; in Figure 19c the nucleus has left traces of a "track" in the cytoplasm. In neither case was there any aster or centrosome apparent. One other egg (Figs. 20a and 20b) shows a condition, probably a later stage of polyspermy, in which there are two sperm nuclei. Both nuclei are in the same condition as the egg nucleus, and entirely separate from the latter and from each other, there being no contact where over-lapping is shown in Figure 20b. A definite funnel-shaped "track" extends from the surface of the egg to this group of vesicles; indeed, a similar appearance is recognizable for a short distance beyond the deepest nucleus (Fig. 20a). I believe the middle vesicle, which is slightly larger than either of the others, is the egg nucleus, and that each of the others is a sperm nucleus, one of which has approached the egg nucleus from either side.

There is, however, no other evidence than that of size and position to show which is the sperm and which the egg nucleus. I have found nothing to indicate what the possible outcome of such a condition would be.

The germ nuclei, whether composed of one or of several vesicles, approach each other, though even at the time of apposition each may be composed of several vesicles. The nuclei shown in Figure 23 as over-lapping are not in contact, a thin layer of cytoplasm occurring between them. The chromatin in the nuclei is in beaded strands, not connected into a network. The nuclei come to lie one against the other, and at the time of apposition may be of equal size (Fig. 24) or unequal (Fig. 25a). The chromatin begins its concentration, preparatory to forming chromosomes, before the membranes of the nuclei disappear (Fig. 25b), and this concentration is first evident on one side of the nucleus. No asters are found in connection with the nuclei at this stage. Whether the definitive chromosomes are formed before the membrane dissolves is uncertain. Figures 24, 25a and 25b show a concentration of the chromatin already in progress, but Figures 26 and 27, which show the first cleavage spindle forming, still show no definite chromosomes, even though in Figure 27 there is only a single vesicle, the sperm and egg nuclei apparently having completely fused. In none of these cases are definite chromosomes present. On the other hand, Figure 23 shows distinct beaded strands in both nuclei, as though the chromosomes were already distinct, though not yet condensed into their definitive form. That there may be a complete fusion of the germ nuclei without any astral figure, seems probable from the conditions seen in Figures 24, 25a and 27, though Figure 26 seems to indicate the presence of two more or less distinct groups of chromatin threads and a spindle already nearly formed.

The cleavage spindles have definite asters, but how these arise I have not been able to determine. We have seen that either the sperm nucleus (Fig. 16a) or the egg nucleus (Fig. 21b) may possess an aster before apposition occurs, but Figures 24 and 25a show the entire absence of asters, as is sometimes the case before apposition. Figures 26 and 27 show two asters in each case, and the beginning of the formation of the spindle, but no hint is given of the origin of the asters.

The conditions described seem to warrant the following conclusions: Spermatozoa usually enter the egg after both polar cells are formed. The penetration may take place at any point on the surface of the egg, but more commonly it occurs in close proximity to the egg nucleus.

Polyspermy is of frequent occurrence. The sperm head begins its transformation into the sperm nucleus soon after entrance, and while close to the surface of the egg. The egg nucleus is often composed of several distinct vesicles, and the sperm nucleus occasionally appears lobed, as though formed by the confluence of several vesicles. An aster may accompany the sperm nucleus, or it may perhaps be in connection with the egg nucleus, but at the time of apposition of the nuclei no astral radiations are present, as a rule. A complete fusion of the nuclei may occur, or they may retain their independence while the first cleavage spindle is forming. Chromosomes seem to take their definitive form only after the first cleavage spindle is present. The origin of the cleavage asters could not be determined.

4. *Cleavage.*— Figure 27, which shows the first-cleavage spindle forming, gives the impression of its formation from the cytoplasm, since the nuclear membrane, still intact, is deeply indented opposite the aster, the astral rays extending into the indentations. The second cleavage spindle (Fig. 28) gives evidence of the same thing, for although the spindle fibres are nearly formed, and the elongated nucleus is in the axis of the spindle, the nuclear membrane is still unbroken. Large conspicuous asters occur at the poles of the second cleavage spindle; the radiations do not enter the large clear centrosphere, nor is there a central body found in it. The chromatin of the nucleus is beginning to produce beaded strands, though definitive chromosomes are not yet formed, and a part of the reticulum remains. In this egg, although the second spindle is forming, the first cleavage furrow is only started, and in other eggs when the third division of the nucleus had begun the first cytoplasmic division was still unfinished, and the second cleavage furrow only just started. This delay in the cytoplasmic division was found by Hargitt (**:04**^{c}) to be common in Pennaria, and we shall see that it is also typical in Tubularia. Further cleavage stages of Pennaria have not been studied.

B. Tubularia crocea.

1. *Oögonia.*— The primordial germ cells divide mitotically to form the oögonia, which are so closely packed around the spadix that their outlines are obscured, if not wholly obliterated, as has been described by Allen (**:00**). The nuclei of the oögonia are relatively large and each contains a single large nucleolus, as Allen has also observed. The nucleolus stains intensely in iron hematoxylin, but in hematoxylin and eosin it selects the acid or plasma stain. Delicate linin fibres

connect the nucleolus with the nuclear reticulum, the chromatin appearing in the reticulum in scattered masses, chiefly at the nodes or close to the nuclear membrane. In preparation for the last oögonial division, the chromatin collects into larger masses, which may become strand like, but do not seem to form a definite spireme. The concentration continues and the chromosomes in the form of a closely packed mass constitute the equatorial plate (Plate **4**, Fig. 29, *o'go*^{2}.). Meanwhile, the nucleolus has disappeared. The last oögonial spindle has no asters, neither could centrosomes be demonstrated, though probably they are present. During the division the chromosomes remain so closely massed that their individuality is entirely hidden. The compact cytoplasm, even before division is complete, may become more distinctly granular and less deeply staining (Fig. 29, *o'go*^{2}.). Some of the oögonia do not undergo this final division with their sister cells, and probably serve to start the next generation of eggs, which develop when the first have formed embryos.

2. *Oöcytes.*— The final oögonial division results in the primary oöcytes. The chromatin of each oöcyte remains for some time in a compact mass (Fig. 29, *o'cy*^{1}.); but after a time it becomes more open. Meanwhile a nucleolus, which selects the plasma stains, has made its appearance (Plate **5**, Fig. 30), and the chromatin mass has opened out into small irregular masses, evidently pieces of the original chromosomes. At this time a differentiation of the oöcytes occurs; some of them at once become young egg cells, while others do not develop further. The latter are present in larger numbers, most of them furnishing food for the growing eggs. In these the chromatin becomes arranged in irregular masses along a delicate linin reticulum, which is limited mostly to the outer part of the nucleus (Fig. 34a). While these oöcytes are perhaps not destined to become egg cells, they do not show degenerative changes (unless they are being, or are about to be absorbed), and hence under favorable conditions they may retain the capacity to form egg cells. In the oöcytes which begin to grow at once the chromatin forms a definite spireme, at first so closely massed as to show little sign of its thread-like character (Fig. 30). As it becomes more open the spireme is easily made out, though I have not been able to determine whether a single thread or several threads are present. Figure 29 shows two oöcytes (*o'cy*^{3}.) already of considerable size, which, from their relative position and from the presence of interzonal filaments, are clearly sister cells. In these the spireme is well shown and each contains a single large nucleolus, though the one belonging to the left-hand cell does not appear in this section.

Before much growth has taken place, the spireme undergoes a further change, the chromatin thread taking on the form of a number of loops (Figs. 31, 32, 33), which, from the first, have a definite polar arrangement; the attached portion of the loops being apparently fastened to the nuclear membrane near a common point, the opposite ends being always closely approximated to the opposite wall of the nuclear membrane, though never attached to it. It is not always possible, however, to demonstrate this condition, since the loops are often long, and extend more than half way around the inner surface of the nucleus. When a number of them cross one another there results such a complicated figure that the polar arrangement, if it exists, is entirely masked, the nucleus apparently containing a long complex spireme. There is little doubt that such a polar arrangement regularly constitutes a stage in the oögenesis of Tubularia crocea. This seems to correspond to the synapsis found in so many other animals, and described by several authors for coelenterates. None of these authors, however, mentions the polar arrangement of the spireme. I could not determine definitely the number of loops present, though in the few that could be counted there seemed to be from nine to eleven.

The further changes of this spireme can be determined only in part. The polar arrangement seems to be lost after a short time and does not appear again during the oögenesis, so far as I could find. Some preparations (Figs. 34, 38) seem to give evidence of a splitting or doubling of the threads composing the loops, but as this appearance has been clearly seen in only a few cases, it may be purely accidental. It would be difficult to conceive that a split condition was represented in Figure 37, which shows a stage of about the same age as that of Figure 38. The spireme continues to get finer and more delicate and also more complex in arrangement, the thread taking on a granular appearance, as shown in Figures 34 and 38. This change to a granular condition continues to such an extent that, in the nearly mature egg, the germinative vesicle shows, besides a nucleolus, only a mass of granules arranged in a very extensive though delicate reticulum, extending throughout the nucleus (Figs. 39–46). As the reticulum is colored by plasma stains more intensely than by nuclear dyes, there results the appearance so characteristic of the germinative vesicle of Coelenterata and some other groups, in which the chromatin is very finely divided and diffused. That the chromatin is present in the reticulum and not in the nucleolus, will be made clear in the following description of the changes which the nucleolus undergoes. This extreme diffusion of chromatin is not always equally marked in different eggs of the same size, the reticulum retaining the deeply staining capacity in some for a long time. Just before the time of polar-cell formation, however, the germinative vesicle shows a minimal affinity for basic stains.

During the changes last described there occurs a great increase in the size of the nucleus. Figures 29–46, all drawn to the same scale, make evident this growth. In the full grown egg the germinative vesicle is, however, very small in proportion to the size of the egg, as may be learned from Plate **7**, Figures 55 (a nearly full grown egg) and 56 (one that has completed its growth). The failure of certain observers to find the germinative vesicle is partly due, without doubt, to its relatively small size and its lack of affinity, at this time, for nuclear stains.

Almost as soon as growth begins, though sometimes much later, the cytoplasm, which at first is compact and finely granular, becomes vacuolated or alveolar, and this condition continues throughout its subsequent development. In the early stages of growth, even when the egg has increased to one-quarter of its final size, no "pseudo-cells" are present. Grönberg (**'98**) described the same condition in young ova of Tubularia coronata, and thought it was an indication that the increase in the size of the ovum was due to the formation of vacuoles in the cytoplasm, no oöcytes, in his opinion, being absorbed until later. But in Tubularia crocea, at least, another explanation is more satisfactory. The oöcytes which are situated near the spadix and often retain their original position till a late stage, are usually the ones which first show signs of growth, and they probably secure nourishment from the spadix. Moreover, in Tubularia crocea, oöcytes are absorbed by the egg during its early growing period, as well as at later stages. The absence of "pseudo-cells" is a sign of complete digestion of absorbed oöcytes, rather than of an increase of the ovum by simple vacuolization of the cytoplasm. Evidence of this is found in the fact that there are oöcytes of many times their original size in which the cytoplasm is still granular or only slightly vacuolated. The presence of "pseudo-cells" is, in part, an indication of the storing of food matter for future use. The absence of "pseudo-cells" is characteristic of the eggs of Pennaria through their entire growth period, as Hargitt (**:00**, **:04**^{c}) has shown; yet during this period the surrounding oöcytes are being absorbed, and they furnish the food for the growing ovum, precisely as in Tubularia.

The oöcyte, though at first nearly spherical or ellipsoidal and having regular outlines (Plate **5**, Fig. 38), later sends out blunt pseudopodia, which may extend for a considerable distance among the oöcytes (Plate **7**, Fig. 55). The boundaries of the oöcytes break down, and their cytoplasm fuses with that of the growing egg, as described by Allen (:**00**), the nuclei of the absorbed oöcytes remaining as the "pseudocells." Some of these remain undigested even in the actinula at the time of its liberation, as Allen showed. The egg as it approaches maturity gradually withdraws its pseudopodia (Fig. 56) and becomes spheroidal. Until this condition is reached the final stages in maturation do not occur. The nucleus, which in the oöcytes is central (Figs. 29–38), remains so for a short time after growth begins, but as soon as pseudopodia are formed, or even before that time, it takes a position at the periphery (Figs. 55, 56).

a. *Nucleolus*.— After the last oögonial division is completed, and while the nucleus is re-forming in the young oöcyte, the chromatin is so closely massed and so intensely stained that nothing can be determined as to the origin of the nucleolus; for when the details of the nuclear structure become visible, the nucleolus is already present.

In the oögonia and young oöcytes the nucleoli and nuclear reticulum stain as follows: with either Ehrlich's or Delafield's hematoxylin and eosin nucleolus red, reticulum blue or purple; with Conklin's picro-hematoxylin the nucleolus yellow or brownish, reticulum blue; with Delafield's hematoxylin and cochineal the nucleolus brownish, reticulum bluish; with Heidenhain's iron hematoxylin both nucleolus and reticulum intensely black or deep blue. In destaining, following the last mentioned method, the nucleolus holds the color as tenaciously as the reticulum. Often fine linin fibres, which stain with the plasma or acid dyes, can be seen extending from the nucleolus to the reticulum.

With the formation of the nuclear reticulum in the oöcyte the nucleolus increases in size, as is to be seen by comparing oögonia with oöcytes (Plate **4**, Fig. 29). Figure 34 (Plate **5**) shows two oöcytes in different stages of development, the older and larger one containing the larger nucleolus. Apparently the increase in the size of the nucleolus is not due to the acquisition of chromatin, since the nucleolus retains its original affinity for acid dyes. Moreover, the spireme itself is at the same time growing in length and volume, and shows an increasing affinity for plasma or acid dyes. Even at the time of synapsis the spireme stains purplish with hematoxylin and eosin, thus selecting some of the acid stain. Hence it seems probable that the increase in the size of both nucleolus and spireme is due to the same cause, perhaps to the absorption of nuclear sap.

The degenerative changes of the nucleolus first become evident in the growing oöcyte when the chromatin has lost its distinctly spireme-like form and is becoming rearranged into a more delicate granular reticulum. In this stage the nucleolus maintains the affinity for dyes that it first had, while the nuclear reticulum behaves as follows: with hematoxylin-eosin the original bluish color becomes purple or reddish; with picro-hematoxylin somewhat less blue, though never yellow; with acid fuchsin-methyl green mixtures the reticulum, at first decidedly green, becomes bluish or even red. At about the end of the growth period, when the nuclear reticulum presents the appearance of finely divided and uniformly scattered granules, the entire nucleus may stain very uniformly. Apparently at this time there is either only a slight chemical difference between the linin and chromatin constituents, at least so far as our stains give evidence, or else the physical condition of the nuclear ingredients is the cause of the uniformity of staining.

The changes which the nucleolus undergoes are either a gradual decrease in size, or a fragmentation, or both. The time at which these changes begin, relative to the size of the growing egg, varies considerably; sometimes they start almost as soon as the egg begins its growth, at other times only at a later period, after considerable growth. Figure 39 shows the nucleus of an oöcyte which has grown to three or four times its original diameter; the nuclear reticulum stains rather faintly; the nucleolus has already decreased in size and is composed of two pieces; in picro-hematoxylin the larger stains yellowish, the smaller bluish, about like the reticulum. Figure 40b possibly represents a later stage or, perhaps, a slightly different method of the breaking up of the nucleolus. Instead of the detachment of one large mass, there has been a separation of several smaller ones. This oöcyte is of about the same size as the one belonging to the nucleus shown in Figure 39, and the staining reactions are the same. The decrease in the size of the nucleolus becomes evident by a comparison with the nucleolus of an adjacent oöcyte nucleus shown in Figure 40a. There are evidences that the nucleolus loses this substance in a liquid form, since the main mass remains of regular shape and sharp outline, while the detached parts suggest, by their form and position, that they are flowing along the network of the nucleus. A larger oöcyte is represented by its nucleus in Figure 41, where, besides a single nucleolus with a vacuole, there are several smaller bodies of nucleolar origin distributed in the reticulum. The nucleus in Figure 42, from an oöcyte grown to one-quarter of its final size, has several similarly staining nucleolar bodies in close contact with one another. Figure

43, the nucleus of a half grown egg, and Figure 44, from the nearly mature egg shown in Figure 56, present about the same conditions. The stains here used, hematoxylin and eosin, gave the small dense fragments in the reticulum a purplish red, and the larger vacuolated portion a bright red appearance. Figures 45, 46 (Plate 6) are from full-grown eggs which had almost entirely withdrawn their pseudopodia; a final fragmentation of the nucleoli is taking place. In one case there is a chain of vacuolated fragments, in the other the fragments are more scattered. Figure 47 is from a nearly full-grown egg, the pseudopodia of which were still extended. All the nucleolar fragments present in the entire nucleus are shown in the drawing, those lying on either side of the section outlined having been drawn as though projected on the plane of that section. This shows the largest number of fragments that I have ever found. The fragments resulting from the breaking down of the nucleolus appear to become incorporated into the nuclear reticulum, though there are probably some portions which are dissolved in the nuclear sap. When the egg has withdrawn its pseudopodia and assumed a rounded form, the germinative vesicle shows no sign of a nucleolus or nucleolar fragments, and since the nuclear membrane has not yet been ruptured, the entire substance of the nucleolus has become disseminated throughout the nucleus.

The nucleolus, and hence the material derived from it, I believe to be non-chromatic, since its staining reaction is different from that of chromatin. The nucleolar substance, however, becomes incorporated into the nuclear reticulum, and perhaps is converted into chromatin. Evidence is not lacking to support this contention. For, with hematoxylin-eosin staining the nucleolus at no stage shows any sign of a blue color; with picro-hematoxylin it is always yellowish, never blue; in the Ehrlich-Biondi acid fuchsin-methyl green combination it is pink and not green, though it is sometimes of a darker pink, and in oögonia is bluish. Further, the fragments of the nucleolus in the nuclear reticulum become changed so as to stain more like the reticulum and less like the nucleolus; in the hematoxylin-eosin stain the nucleolus is red, the fragments in the reticulum purplish; in fuchsin-methyl green the large nucleolus is red, but the smaller fragments in the reticulum vary in color, some being red, others blue and still others green. This seems to me to indicate a rather gradual chemical change in the substance of the fragments. In picro-hematoxylin preparations the larger pieces are yellow, the smaller ones blue.

To sum up: the nucleolus during the growth period of the egg seems to be composed of non-chromatic substance. It disappears in the germinative vesicle at the end of the growth period before the dissolution of the nuclear membrane takes place, part of it entering the reticulum and part, perhaps, the nuclear sap. While not chromatin, it appears to be capable of transformation into chromatin, and so may perhaps serve as a storehouse for material needed in the formation of the chromosomes. Its disappearance is by a loss of liquid substance or by fragmentation, or by both.

3. *Polar-Cell Formation and Fertilization.*— Just before the time of polar-cell formation the egg is rounded and without pseudopodia; the germinative vesicle lies close to the periphery of the egg, its nucleolus has disappeared and its chromatin is finely divided and distributed along the nuclear reticulum. A decrease in the size of the germinative vesicle takes place, as a comparison of Figures 45 and 46 with Figure 48 shows.

Beginning at this time, or somewhat earlier, the reticulum of the germinative vesicle again stains intensely with nuclear dyes. The chromatin is assembled at the nodes of the network in masses, usually irregular in shape and granular in appearance, which suggests the grouping of smaller granules into the larger mass. The appearance in the fixed egg is well shown in Figure 48, though the germinative vesicle at this stage is usually not so far from the surface of the egg. Often the cytoplasm around the nucleus is finely granular and destitute of the vacuoles which are present in the rest of the cytoplasm.

The history of the subsequent changes has been made out in part only, since certain stages could not be found. The most characteristic condition found when the time of polar-cell formation approaches is that shown in Figures 49 and 50. The nucleus has become ovoidal, the long axis having a radial position. The outer end of the nucleus is often sharply pointed, and this seems to represent a later stage than the more rounded condition shown in Figure 49. Almost always at this stage radiations are present at the outer end of the germinative vesicle, and the surface of the egg itself is often raised into a more or less conical elevation at this point. The centre of the radiations is always between the germinative vesicle and the surface of the egg; in no case were centrosomes found, nor were radiations seen at any other part of the nucleus. Since polar cells were apparently not present, it seems probable that this is a prophase of the first maturation figure, though a somewhat similar condition in Pennaria (Figs. 14, 15) represents a stage after the first polar cell has been formed. There is no evidence upon which to determine whether there is a division of this aster to form two centres, or whether the spindle centres arise in a different place and manner, no stages having been obtained connecting this with the maturation spindle. In Tubularia, the oöcytes not used as food often remain in contact with the egg, and when degenerating they bear so close a resemblance to polar cells as to make it almost impossible to distinguish between the two, unless the polar cells are still actually joined to the egg. However, what little evidence there is favors the view that this nuclear condition in Tubularia is a stage immediately preceding the formation of the first polar cell. A fragmentation of the nucleus at or before this time, as claimed by Allen (:00), does not occur, the polar cells being formed without doubt by mitosis. There is no case in which I have not been able to find a nucleus of some sort in each egg examined, and never has there been the slightest sign of fragmentation. A spindle found close to the surface of the egg in a nearly radial position (Fig. 51a) may represent the first maturation spindle. Centrosomes and asters could not be detected. While the preservation of this egg was not perfect, it was good enough to show the structure of the spindle itself clearly. The chromosomes in the equatorial plane, while very small and massed together, show a splitting, and the two parts are just beginning to separate. The small sphere outside the egg, at the left of the figure, may represent a polar cell, but in size and general appearance it is more like a degenerate oöcyte. In the centre of this egg was found another nuclear body (Fig. 51b), which probably represents the sperm nucleus. The second maturation spindle was not seen in Tubularia.

Figure 52 shows the two polar cells, one of which is still joined to the egg by interzonal filaments (the body shown with dotted outlines was in the section following the one drawn with a continuous outline). The two polar cells are equal in size, each contains some cytoplasm, and the chromatin is in several masses not surrounded by a membrane. The egg nucleus has the form of a single spheroidal vesicle with the chromatin scattered along a reticulum; no asters were present. Figure 53 shows a nearly tangential section of another egg, provided with two polar cells outside the membrane. In this case, too, the chromosomes are aggregated into several discrete masses. The egg nucleus occupies a superficial position in an elevation of the surface of the egg, and there are no cytoplasmic radiations in its vicinity.

In one case only (Fig. 54) was a spermatozoön found within the egg. The sperm head had as yet undergone no marked change. It was accompanied by a distinct aster, in which, however, no centrosome could be detected, though several stains were used in an attempt to demonstrate one. Unfortunately the condition of the egg nucleus could not be determined. The only evidence of such a body was an aster in the cytoplasm not far from the spermatozoön and near bodies which, though staining rather faintly, appeared to be chromatic. Their condition and arrangement could not be made out with sufficient clearness to determine whether or not they were definitive chromosomes. The surface of the egg was elevated at this point, and it may be that a polar cell was about to have been formed. In the vesicular sperm nucleus represented in Figure 51b, the chromatin is scattered in granules through the reticulum. No asters or radiations of any sort accompanied it. At the surface of the egg there still remained a small cone, but between it and the sperm nucleus there was no "path" in the cytoplasm. Since this egg had a maturation spindle, it seems probable that the spermatozoön may enter before the polar cells are formed. No further stages in fertilization were found, the next more advanced stage observed being that of the first cleavage spindle in metakinesis.

4. *Cleavage.*— Segmentation is total but unequal, and it is often irregular in its progress. There is no localization of yolk material to account for this irregularity, which may possibly be due in part to the fact that development takes place in a closed gonophore; but this will not wholly explain the irregularity, for the segmentation of Pennaria tiarella as described by Hargitt (:00, :04^c) is strikingly irregular, and yet this egg develops free in the water. Colonies of Tubularia crocea collected from different localities, or from the same place at different seasons, show in some cases quite regular cleavages; other lots are almost as constantly very irregular. Whether there is any significance in the fact that colonies collected in the fall were more regular in cleavage than those collected from the same point in the spring, I do not know; but this difference becomes evident upon comparing series of sections of two such lots. It may be stated as a further general conclusion, supported by all the eggs examined, that cleavage is always accomplished by mitosis. This is especially evident in the early stages, just where Allen (:00) claims to have found indications of the reorganization of a previously fragmented nucleus. It was because of this contention of Allen that especial care was taken to examine closely the early cleavages, and in no instance was there any indication whatever that a reorganization of fragmented particles was occurring. Furthermore, all cleavages up to the end of segmentation showed no sign of amitotic division, it being possible in almost every case to determine the line of descent of each nucleus through preceding mitoses.

A certain polarity is shown by the egg, though its poles are quite variable in their relation to the spadix of the gonophore. Reference has already been made to the difficulty of finding maturation spindles and polar cells, and to the close similarity between the latter and degenerating oöcytes. These conditions contribute to the difficulty of determining the poles of the egg. The relation of the germinative vesicle to the spadix is shown in Figures 55, 56 (Plate 7), which represent nearly mature eggs in which the germinative vesicle lies at the periphery and probably marks the point where the polar cells would have been formed. In all eggs which I have examined the germinative vesicle, just previous to polar-cell formation, is on the side of the egg which is opposite the spadix, and this holds good for the position of such polar cells as I have seen; but no more definite relation between the spadix and the animal pole of the egg could be determined. As will become clear during the description of the later stages, these facts are in harmony with cleavage conditions. For instance, during the early cleavages the nuclei are always on the side of the egg opposite the spadix, and cytoplasmic division seems to start in this region. We are justified, then, in assuming a more or less definite polarity for the egg, as Allen (:00) earlier pointed out, and as others have observed in the eggs of coelenterates. It seems to me, however, that a comparison of the germinative vesicles in growing eggs warrants the conclusion that their position in relation to the spadix is not definitely fixed, and therefore that the exact position of the pole is variable.

a. Early Cleavage.— Figures 57a and 57b show the nuclei of an egg just after the first cleavage. Interzonal filaments are present and the reorganization of the chromatic substance has resulted in a double vesicle in each case. From conditions found in other nuclei, it is very probable that these would have fused into a single vesicle. Figure 57a shows one polar cell, which probably still occupies the animal pole of the egg.

Figures 58a and 58b show the second nuclear division completed, from which it is clear that the division of the two nuclei has been almost perfectly synchronous. Figure 58c, a portion of figure 58b more highly magnified, shows the appearance of the interzonal body, of the remains of the interzonal filaments, of the nucleus reorganized into a single vesicle and of the second cleavage furrow at the beginning of its formation. That the division of the cytoplasm lags behind that of the nucleus, is evident from an inspection of Figures 58a and 58b, the first cytoplasmic division having proceeded only a short distance into the egg, even after the second nuclear division. This condition is the usual one, for in no case in the early cleavage stages have I found the cytoplasmic division completed before the next nuclear division started. This delay in the cleavage of the egg is so extreme in some instances that cytoplasmic division does not begin till a number of nuclei are present. Similar conditions are common in other coelenterates, and have already been indicated by other observers.

The result of the first and second cleavages may be respectively two and four equal blastomeres, as is foreshadowed in Figure 58. The eggs which I have seen at the end of the first cleavage, in cases where the division of the egg followed immediately on that of the nucleus, showed approximately equal parts; but the second cleavage may result in the formation of very unequal parts, and from this time on inequality is the rule. I have no doubt that the first division may also sometimes result in unequal blastomeres. Figure 59 illustrates the inequality in the size of the blastomeres resulting from the second cleavage.

b. Later Cleavage.— Usually the cleavage planes for a considerable time correspond to meridional divisions in eggs that segment more regularly. An equatorial division rarely occurred before the 12-cell stage, and usually it was much later in making its appearance. Sections from eggs with 20–30 nuclei each are shown in Figures 60–62; the meridional furrows, the delay of the equatorial cleavage and the polarity of the egg, all being well shown. The elongated shape of these eggs is probably due to external causes, the development of several eggs in one gonophore having led to a compression which has influenced the direction of growth, and may have helped to determine the position of the planes of cleavage.

Traces of segmentation cavities are seen in Figures 60–62, and such cavities, I believe, usually occur. In this I must differ from Allen (:00), who did not in any instance find a cleavage cavity in Tubularia. It is clear from these sections that the cleavage cavity is not a typical one, such as we know in eggs of echinoderms, for instance. Several separate spaces occur between adjacent cells, beginning as early as the 8-cell stage. It is doubtful whether these spaces unite to form a single cavity in all cases, perhaps they do not even in the majority of cases; there is no question, however, that a single cavity does arise in some instances. For example, Figure 63 (Plate 8), a section of an egg with 40–50 cells, shows a rather irregular, but single segmentation cavity, which is for the most part surrounded by a single layer of cells. This stage, I believe, corresponds to the typical blastula of other animals, from which it differs chiefly in its irregularity of form. That this stage, which marks the end of segmentation, does not always result in a blastula of precisely this character is probably due, in part at least, to pressure consequent upon development within a closed gonophore. The further division of cells accompanies the process of the formation of the germ layers.

The proliferation of nuclei, not followed at once by cytoplasmic division, was somewhat more common in eggs collected in the spring, though this probably has no especial significance. In this segmentation there were no signs of nuclear reorganization from a previously fragmented nucleus, and no signs of amitosis, the evidences of nuclear division always pointing to mitosis. Figures 64 and 65 are outlines of two different eggs with all nuclei projected on the same section by the superposition of carefully made camera drawings. Four pairs of nuclei are shown in Figure 64, and their origin from a single nucleus is not hard to conceive. The polarity of the egg previously noted is well shown here, the nuclei being located on the convex side of the egg, the one opposite the spadix. A polar cell (*cl. pol.*) is shown and the cleavage of the cytoplasm has begun, one furrow having separated off a small blastomere (shown by dotted lines), and another furrow having started from the region of the polar cell. In Figure 65 fourteen nuclei are shown; one blastomere is entirely separated and a second furrow has made its appearance. The absence of polar cells makes an exact orientation of the egg impossible, though the aggregation of the nuclei on the side opposite the spadix is nearly as pronounced as in the previous figure.

A rather late stage of cytoplasmic division is shown in Figure 67, though there are still many more nuclei than blastomeres, and the inequality of the blastomeres is very evident. In Figure 68 is shown a still later stage, in which the cells are more nearly equal, though variations in size still occur. In such cases the indications of a cleavage cavity are not so frequent; still, as Figure 67 indicates, a small cavity does occur. In Figure 68 there is no cleavage cavity, merely a solid mass of cells, but it is not impossible that at an earlier stage this, too, had a cleavage cavity, which became obliterated during subsequent cleavages. The embryos resulting from this kind of segmentation apparently differ in no way from those formed by the first method described. Figure 66 shows a section of an egg from the same hydranth as that of Figures 64 and 68; here the nuclear divisions have been followed at once by cleavage of the cytoplasm.

c. The Formation of the Germ Layers.— The stage shown in Figure 63 is near the beginning of the process of delamination which ends in the separation of entoderm from ectoderm. At this stage several entoderm cells have already been cut off, and the radial position of some of the spindles clearly shows that this number will be increased by the approaching divisions. Figures 69 and 70 represent more advanced stages in this process, Figure 70 especially showing that the entoderm cells are formed by a "multipolar delamination." The ultimate result of this process is the formation of a solid mass of cells which, in this and other coelenterates, has often been called a morula, and considered to mark the end of segmentation. It seems clear that in Tubularia crocea, where a true blastula occurs, this process should be regarded as a part of the germ-layer formation. The cells composing the layers of this solid mass, it is true, do not assume their definitive positions and relations until somewhat later, as the result of further divisions and rearrangement; but that the separation of entodermal cells has already begun cannot be doubted. During the formation of the germ layers the interzonal filaments of the mitotic figures persist for a long time (Plate **9**, Figs. 73, 74, 77), usually being still evident when the next division begins. This greatly facilitates tracing the line of descent of each nucleus, and shows that nearly up to the planula stage, at least, all divisions are clearly by mitosis.

The deeply cleft or bilobed condition shown in Figure 70, which is sometimes met with, is strongly suggestive of a double blastula. A comparison with other somewhat similar stages leaves the impression that this may be the result of an uncompleted first cleavage division; it is as though the first division plane got but little beyond the condition shown in Figure 58 (Plate **7**), and that then each half continued to segment independently. It is not impossible that in some cases this is due to a mechanical restraint causing a bending in of one side. The cleavage cavity is common to both halves, and entoderm cells are being cut off from the deep ends of the blastula cells. It does not seem probable, however, that such a blastula gives rise to a twin or double embryo, for no embryos of that sort were ever found.

d. Double Nuclei.— In some of the cleavage stages the nuclei are distinctly double and closely resemble conditions found by Häcker (**'95, :03**) in copepods. In Tubularia this is very rare in the early cleavages, one case only (Plate **7**, Figs. 57a, 57b), at the end of the first cleavage, showing more than a single vesicle. The cells of the blastula stage, however, and of the embryo during the formation of the germ layers as a rule have the nuclei double; indeed all the eggs of this stage which were examined showed this condition in some blastomeres. While not all nuclei presented this appearance, the majority were double and the apparent singleness in others was due, in some cases at least, to the fact that the line of division between the halves was parallel to the plane of the section. There is no difference in this regard between the nuclei of cells destined to form ectoderm, and those which are to form entoderm.

Each resting nucleus typically consists of two vesicles (Plate 9, Figs. 71–73), each of which contains one or more nucleoli and a network in which the chromatin is arranged. That the halves are entirely distinct, is sometimes very plainly shown (Fig. 75), especially when the vesicles are so oriented that the plane of their contact is perpendicular to the plane of the section. These nuclei have arisen by mitosis, as the interzonal filaments of Figure 73 clearly indicate, and later they divide by mitosis (Fig. 74). The chromosomes arise independently, but synchronously, in the halves (Figs. 75, 76), though it is often impossible to determine whether they remain separately grouped after the dissolution of the nuclear membrane. As soon as they enter the equator of the spindle, they become so massed together that their individuality is hidden, and polar views of the spindle in this stage often show the chromosomes in a single mass. On the other hand, sections of the spindle when seen in polar view sometimes show the chromosomes still arranged in two groups (Figs. 79a–79c); while in these cases the individual chromosomes are not distinct, their arrangement in two groups is unmistakable. These figures are especially selected to show the double character which the chromosomes sometimes exhibit; but many other spindles in similar stages when seen in polar view do not show a double grouping of the chromosomes. Hence, the double nature of the nucleus may not always manifest itself in the spindle stage. In the reorganization of the chromatin to form the daughter nuclei, however, two vesicles were almost invariably found (Fig. 77), and these, by an increase in size, assume the condition characteristic of the resting stage (Figs. 71–73).

When the cells of the germ layers are beginning to take their definitive positions and arrangement their nuclei usually appear single, but two nucleoli are generally present. By the time the ectoderm cells have attained the size and arrangement found in the planula and are separated from the entoderm cells by a supporting membrane, the nuclei of both layers have entirely lost the evidence of their double nature, each nucleus being spherical, and containing a single nucleolus. Double nuclei are entirely lacking in the planula and actinula and were never found in the cells of the gonophore or in the primordial germ cells.

To recapitulate: segmentation is total, and may be nearly equal or decidedly unequal, and it may be more or less irregular. All cell divisions, at least up to the formation of the planula, take place by mitosis. The egg presents a rather marked polarity. Segmentation of the egg may follow nuclear division almost at once, though always slightly delayed, or nuclear proliferation may continue for some time before cytoplasmic division begins. A segmentation cavity usually occurs, though it may be represented by separate spaces between adjacent blastomeres. A nearly typical blastula — a single layer of cells surrounding a large segmentation cavity — is sometimes produced, but this may be more or less modified, even to the complete obliteration of the cleavage cavity. In some cases no stage exactly comparable to a blastula can be discovered. The solid mass of cells, the so-called "morula," which sooner or later results, is to be regarded as the outcome of the process of germ-layer formation and not as the end of segmentation. The germ layers are the result of a primary multipolar delamination, though the definitive ectoderm and entoderm are later more completely differentiated by a further division and rearrangement of the cells of the early germ layers.

Double blastulae have been found, which are probably the result of an incomplete first cleavage. The nuclei of early cleavage stages are usually single, those of blastulae usually consist of two vesicles. Chromosomes arise independently but synchronously in the two parts of the double nuclei, and their distinctness may or may not continue throughout the mitosis. Daughter nuclei take the form of two apposed vesicles which, however, remain distinct through the entire resting period. When the germ layers have been definitely formed and are separated by the supporting membrane, double nuclei no longer appear.

IV. Discussion and Historical Review.

1. *Growth of eggs.*— It is well known that in the Coelenterata, although the primordial germ cells are all alike, only a few of the many oöcytes become mature, the rest serving as food for the survivors. Most authors have believed that the circumstances of favorable position and nourishment determine which oöcytes become egg- and which food-cells. Brauer (**'91**^{b}) and Wulfert (**:02**), however, believe that a differentiation occurs during the migration of the germ cells into the gonophore, though the latter maintains that this early differentiation is the result of favorable position and nourishment. Grönberg (**'98**) and Müller (**:08**), on the other hand, think the differentiation occurs in the oöcytes.

In Tubularia crocea the oögonia are all alike; but in the oöcytes, even before growth begins, a difference is apparent, the chromatin in the nucleus being either (1) in the form of a spireme or (2) scattered in grains along a network. The oöcytes which possess a nuclear spireme begin growth at once, the most of those with a diffuse network serve as food, though if they escape this fate they seem to be able later to form egg cells. The cause of this differentiation seems to be better nourishment, as the oöcytes near the spadix are most often the ones which first begin to grow.

The cell which begins to grow absorbs the surrounding cells by one or another of various methods. The nucleus of this growing oöcyte is commonly believed to become the germinative vesicle, while the nuclei of the absorbed cells are used as food at once, or remain in the cytoplasm as yolk bodies, "pseudo-cells." However, Allen (:**00**, p. 300) says in regard to Tubularia (Parypha) crocea: ". . . . the nuclei of the growing cells disappear at an early stage, so that only the nuclei of the smaller cells persist." "It thus becomes impossible to tell which is the controlling cell." This is exactly the opposite of what I find in the same species, for in my preparations the nucleus of the growing oöcyte is always present and easily distinguished, because of marked differences between it and the degenerating nuclei of the absorbed oöcytes.

2. *Synapsis and Reduction.*— In Tubularia after the last oögonial division, and before the growth of the primary oöcyte begins, the chromatin of the nucleus is in the form of a spireme, which soon takes the form of loops showing a definite polarity, the open ends of the loops apparently being attached to the nuclear membrane. This condition seems to correspond to similar polar arrangements of the spireme in the synapsis stage of other metazoa. A synapsis in Coelenterata has been described in spermatogenesis by Guenther (:**04**) and Downing (:**05**) for Hydra, and by Bigelow (:**07**) for Gonionemus. Stschelkanowzew (:**06**) figures nuclei of spermatocytes in which the chromatin appears to be in a contraction stage or polar arrangement, but does not so describe it. The characteristic condition is such as might result from a contraction into a dense mass. Bigelow thinks that in Gonionemus this is artificial. Trinci (:**07**) has described a synapsis as occurring during the oögenesis of several Hydromedusae. He also shows a contraction phase, though in his figures the chromosomes still show a rather definite polarity after the contraction.

After the contraction Guenther found a reduced number of chromosomes, while Downing thinks the reduction occurs in the telophase of the last spermatogonial division. Bigelow (:07, p. 371) is of the opinion "that a pairing of individual chromosomes does not occur at all in Gonionemus, but that synapsis occurs between the chromatic microsomes; and takes place while these are intimately associated in the homogeneous net" of the primary spermatocytes. A. und K. E. Schreiner (:06–07) in attempting to correlate the synapsis stage in all animals, have claimed that during the polar arrangement of the loops, at first present in the same number as the somatic chromosomes, a reduction in number to one-half takes place by a side to side conjugation. This is apparently not true for many animals, where an end to end conjugation occurs. The polar loops were not commonly observed in Tubularia, and their arrangement was not always clear. In some cases the number of loops was about half, in other cases more than half, the number of chromosomes in somatic cells. A reduction is thus suggested as taking place during this period, though the evidence is not sufficient to furnish convincing proof; and no evidence could be obtained as to how the reduction occurred.

Trinci (:07) found that after synapsis a continuous thread was present in Tiarella. This segmented into chromosomes,— though he does not say whether or not in a reduced number,— which anastomosed into a network persisting throughout the growth period. In Phialidium the chromosomes, formed in the same way, scatter through the nucleus and lose their staining capacity, but retain their individuality through the growth period. In Tubularia I find that the loops soon lose their polarity, and become more delicate and distinctly granular; in a few preparations threads lying side by side suggest a splitting of the loops, but the evidence on this latter point is so meager that it may be without significance. The chromatin in the germinative vesicle of eggs in later stages of growth is always finely granular and scattered along the reticulum, and all trace of the loops of the synapsis period is lost.

The formation and behavior of the chromosomes in the maturation spindle I could not observe in Tubularia, and the important evidence which this would offer in regard to reduction is therefore lacking. It was possible, however, to observe these stages in Pennaria, and the conditions there may help to fill the gap in the history of Tubularia. In the equatorial plate of the first maturation spindle of Pennaria the chromosomes were present in the reduced number. Furthermore, they were partly in the form of tetrads, and in later stages in the form of x- and v-shaped figures; this suggests a splitting in the heterotypical fashion, such as is common in maturation mitoses. Since in the later growth period the chromatin was present in the form of granules (as in Tubularia) diffused through the nucleus, and a spireme or definitive chromosomes were always lacking, it seems clear that the reduction must have occurred during early growth, or before; the conditions in Tubularia suggest that this happened in the oöcyte just previous to the growth. The reduction process was consequently not fully worked out in either Pennaria or Tubularia alone, but in view of the close relationship of these genera and their similarity in many points during oögenesis and development, it seems not entirely without justification to consider the process determined in part from each as giving an approximately correct picture of the whole matter in both genera.

Tetrads in the reduced number have been found in the first maturation spindle of Tiara (Boveri, '90), Clava (Harm, :02) and Cunina (Stschelkanowzew, :06). In Linerges Conklin (:08) finds tetrads, but says nothing concerning reduction, while in Gonothyraea Wulfert (:02) found a reduced number of chromosomes not in tetrads. These forms, then, as well as Tubularia crocea and Pennaria tiarella, give evidence of a reduction occurring before the maturation spindle forms, and since the germinative vesicle contains diffuse granular chromatin during the growth of the egg, the reduction presumably takes place before growth begins. The Hydromedusae thus seem to agree with other Metazoa in the time of reduction, but the manner of reduction remains undetermined.

3. *Polar-cell Formation.*— In a number of Hydromedusae, including Pennaria and Tubularia, the germinative vesicle has been thought to fragment and entirely disappear about the time polar cells should be formed. Allen (:00, p. 303) says regarding Tubularia (Parypha) crocea: "I. . . .am forced to the conclusion that the nucleus of the mature egg is formed by the reorganization of the fragments of the nuclear matter scattered through the cytoplasm." Hargitt (:04^a, :04^c, :06) thinks that there may be sometimes in Pennaria, Tubularia mesembryanthemum and other forms an apparent dispersal of the chromatic matter of the germinative vesicle throughout the cytoplasm, and Müller (:08) maintains for Margelopsis and other species that the disappearance of the germinative vesicle is "absolutely complete." As this author finds both chromatin bodies and maturation spindles, he apparently does not refer to an actual dispersal of the chromatin.

In every egg of Pennaria and Tubularia which I have examined, I have found the nucleus in some phase of its cycle. It is often very faint, and is usually not accompanied by any marked concentration or specialization of the cytoplasm; but no sign of its fragmentation has ever been seen. The supposed disappearance of the germinative vesicle at this time, I believe to be due simply to the usual dissolution of the nuclear membrane and the mingling of karyoplasm with cytoplasm which is characteristic of the prophase of mitosis. After this dissolution of the nuclear membrane maturation spindles are formed, two polar cells are constricted off from the egg, and the chromosomes remaining in the egg form a definite vesicular egg nucleus.

4. *Nucleolus.*— The nucleolus of oöcytes of Hydromedusae has been described as plasmatic and not aiding in the formation of chromosomes by Brauer ('91[a]), Häcker ('92), Morganstern (:01), Wulfert (:02), Harm (:02), Trinci (:07), Müller (:08), and Conklin (:08). Trinci (:05) finds in the Eucopidae that the nucleolus divides into pieces, some of which are chromatic and resemble "pseudo-nucleoli" of Amphibia; in Phialidium the same author (:07) describes a chief nucleolus, with acid and basic constituents, not aiding in the formation of chromosomes, and smaller chromatic nucleoli, which help to form chromosomes. Stschelkanowzew (:06) maintains that in Cunina the chief nucleolus alone, which is chromatic, forms the chromosomes, though small plasmatic nucleoli are also present. Bigelow (:07) describes the chief nucleolus in Gonionemus as staining like chromatin, but it takes no part in chromosome formation and is considered as a by-product; accessory nucleoli which occur are plasmatic bodies. The nucleoli which do not form chromosomes are either (1) cast into the cytoplasm, where they are dissolved,— Brauer ('91[b]) Tubularia mesembryanthemum, ('91[a]) Hydra (in part), Häcker ('92) Aequorea, Morganstern (:01) Cordylophora, Harm (:02) Clava, Hargitt (:04[a]) Pachycordyle, (:06) Clava, Trinci (:07) Tiarella,— or (2) fragmentation occurs and the pieces are for the most part dissolved in the nucleus — Brauer ('91[a]) Hydra (in part), Wulfert (:02) Gonothyraea, Trinci (:05) Eucopidae (in part), (:07) Phialidium, Müller (:08) Cladonemidae and Codonidae.

In Pennaria and Tubularia crocea the nucleolus is entirely plasmatic in all oögonia and oöcytes. In Pennaria it seems gradually to dissolve in the germinative vesicle, without fragmenting. In Tubularia it may lose some of its substance in liquid form, or in fragments, at an early and variable stage in the growth of the oöcyte; but eventually the remainder fragments into few or many parts. Of these, some may dissolve in the nuclear sap; others are added to the nuclear reticulum and are apparently transformed into chromatin. The complete disappearance of the nucleolus within the germinative vesicle before the dissolution of the nuclear membrane, and before the chromosomes are formed, suggests the possibility that this body may contain substances necessary to the formation of the chromosomes, and if so, is not superfluous matter nor a by-product.

The opinion expressed by Bigelow (:**07**, p. 367) that "there is. . . . no conclusive evidence that the chief nucleolus in invertebrates ever normally contributes to the formation of the chromosomes of the first cleavage spindle," is not borne out by conditions in Cunina (Stschelkanowzew, :**06**), or in Tubularia crocea. And as regards other invertebrates, Jordan (:**08**[a], :**08**[b]) has shown that the nucleolus of Echinaster fragments, and that from these fragments alone the chromosomes of the maturation spindle are formed; further, that in Asterias the nucleolar substance aids in forming the chromosomes. It seems to me that the claims of Rohde (:**03**) as to the close relationship existing between all nucleoli and chromatin, which Bigelow says (p. 365) "are, in the main, supported by the conditions in Gonionemus," furnish presumptive evidence that the nucleolus may contribute to the chromosomes. If all nucleoli arise in the nucleus from chromatic microsomes, directly or indirectly, as Rohde claims, why may they not return to their original condition as chromatic bodies? This appears to be exactly what happens in Tubularia, where the nucleolus dissolves or fragments entirely within the germinative vesicle, and the pieces become incorporated in the nuclear reticulum. As R. Hertwig ('**98**, p. 713) long ago said: ". . . . in the dissolution the material of the nucleolus unites with the chromosomes," and in its new formation the nucleolus comes from the chromatin. Hertwig considers the nucleolar substance to act like a cement in uniting the chromatin into chromosomes. He says (p. 714) further, ". . . . between plastin- and chromatin-nucleoli, indeed, no sharp boundary exists. . . . . between both forms of nucleoli transitions exist, and one can arise from the other." Ruzicka (:**06**, p. 556) has summed up the matter thus: "The nucleolus can change itself into either nuclear substance, or cell body, or spindle, and can again reform from these conditions."

5. *Cleavage.*— Allen (:**00**) states that the first nuclei of segmentation in Tubularia crocea appear as small masses reorganized from a previously fragmented nucleus, and she found the earliest mitosis occurring when four nuclei were present. Hargitt (:**04**[c], Pennaria) (:**04**[a], Tubularia mesembryanthemum and other forms), Hickson ('**88**, '**90**, '**94**, Hydrocorallinae) and Hill (:**05**, Alcyonium) all speak of a nuclear fragmentation and a later reorganization of the fragments to form cleavage nuclei. In the specimens of Pennaria and Tubularia crocea which I have examined, there was no sign whatever in any egg

of fragmentation. Moreover, I have been able to establish a direct descent of the cleavage nuclei of the blastomeres from the germinative vesicle of the growing oöcyte, through the maturation spindles, the germ nuclei, and the first segmentation nucleus, each division being by mitosis. No step is lacking, and in Tubularia I have traced this line of descent into the nuclei of both ectoderm and entoderm of the planula, and have found each nucleus arising by a typical mitosis.

Brauer ('91^b) distinguished two methods of segmentation in Tubularia mesembryanthemum: (1) a regular segmentation of the egg following nuclear division, and (2) a nuclear proliferation not immediately followed by the differentiation of cells, this taking place only at a later period. Hargitt (:04^a) showed that while such differences exist, there is no sharp distinction between them. Allen (:00) found essentially the same conditions in Tubularia crocea, and my observations are confirmatory, though I often found in the cleavage a regularity which she never observed. As has often been found in Hydromedusae, the cleavage of the egg is delayed, and any nuclear division may be under way, or even finished, before the egg has completed the segmentation corresponding to the immediately preceding nuclear division.

In the late development of the egg a solid mass of cells is characteristic of nearly all Hydromedusae. This has been interpreted in two ways: Conn ('82), Allen (:00), Harm (:02), Hargitt (:04^a, :04^c), Stschelkanowzew (:06), Brooks and Rittenhouse (:07) think this represents the end of segmentation, no cleavage cavity having been formed, i. e., the solid mass of cells is a true morula. On the other hand, Claus ('82), Hamann ('83), Merejkowsky ('83), Brauer ('91^a, '91^b), Gerd ('92), Häcker ('92), Bunting ('94), Morganstern (:01), Wulfert (:02), Tannreuther (:08), and others have found a cleavage cavity during the course of segmentation, and consider that a blastula is formed, which represents the end of segmentation. The solid mass of cells, according to this view, is not a true morula, but in part a result of the formation of the germ layers.

The conditions found in Tubularia crocea agree with the second view, viz: that there is a blastula, but not a true morula. That a blastula is sometimes formed in Tubularia crocea admits of no question, as some of my figures show. However, there is as little question that the segmentation cavity may be reduced to more or less separated spaces between the cells, and possibly may sometimes be entirely lacking. This reduction of the segmentation cavity may be due, in part, to the fact that the development takes place within a closed gonophore; but there seems also to be a tendency to an abbreviation in this stage of the development. This abbreviation is most evident when the cleavage cavity is greatly reduced or lacking, in which case cleavage and germ-layer formation may not be sharply separated from each other, as Wulfert (:02) also found in Gonothyraea.

By a division of the cells of the blastula, the cleavage cavity becomes entirely filled with cells. This happens by the process designated by Metschnikoff ('86) as primary delamination of the multipolar sort. The same thing was found to occur in Tubularia mesembryanthemum by Brauer ('91) and by other workers on other Hydromedusae. Since a blastula stage does occur, this part of the development clearly belongs to the germ-layer formation, the inner cells representing primary entoderm and those at the surface primary ectoderm; a true morula, therefore, does not occur. The definitive ectoderm and entoderm are formed by a further division, specialization, and readjustment of the primary ectoderm and entoderm cells, and the eventual formation of a supporting lamella between them.

This interpretation is not at all inconsistent with the syncytial conditions described by Hickson ('90, '94), Hargitt (:04^{a}, :04^{c}), and Brooks and Rittenhouse (:07). Indeed, from the figures of the last two authors it appears that the syncytium represents the so-called morula, and that an earlier primary delamination has occurred. Their figures also show between the cells spaces, more or less connected, which are indications of a reduced segmentation cavity. The very great delay in the formation of definite cells in Eudendrium (Hargitt, :04^{b}) and the Hydrocorallinae (Hickson, l. c.) is due, as these authors state, to special conditions of development, viz: the presence of yolk, and development within a very narrow cavity, from which the embryo must later migrate.

6. *Double nuclei.*— Multivesicular nuclei have been found in many animals, chiefly in the re-formation of the daughter nuclei after mitosis. Often each chromosome occupies at first a separate vesicle, but these commonly fuse into a single vesicle, which is characteristic of the resting condition. Such nuclei have been found in Hydromedusae by Metschnikoff ('82), Wulfert (:02), and Hargitt (:04^{c}).

Distinctly double nuclei, similar to those found in the cells of the blastula and later stages in Tubularia, have been less often described. In Coelenterata only Conklin (:08) found sometimes in Linerges in the first division such double vesicles, which he thinks represent the halves of egg and sperm nuclei. Häcker ('95) and Rückert ('95) were the first to describe such nuclei, finding them in Copepoda.

They found the resting nuclei double, and sometimes the spindle was double and the chromosomes in two distinct groups. They believed there were thus represented in the cleavage nuclei maternal and paternal elements which remained distinct. Conklin (**:01**) gave the same interpretation to double nuclei found in Crepidula during the telophase of mitosis, and sometimes continuing throughout the resting period. The nuclei were typically composed of two vesicles, each with a nucleolus, though the vesicles and nucleoli sometimes united and thus became single. This condition he found in nearly all stages up to that of 60-cells. Häcker (**:03**) carried his earlier investigations on Copepoda further, and described the separateness of the two portions of the nucleus in all stages of rest and mitosis, from the fertilized egg to the germ mother cells. In Triton, Rubaschkin (**:05**) never found two nucleoli in any resting nucleus; but in early cleavages he occasionally found double nuclei in resting stages, though not during mitosis. In the 16-cell stage and later (i. e. about the blastula stage) he found two vesicles in the resting stage. During mitosis the spiremes formed synchronously in the two vesicles, and for a short time after the nuclear membrane was lost the chromosomes remained in two ill-defined groups; but in the equatorial plate they constituted a single mass. In some blastulae he found no double nuclei in any cell, and in others there were three vesicles, which remained distinct for as long a time as double nuclei did. Because of the equality of the vesicles in size, and their synchronistic differentiation, he believes they represent maternal and paternal constituents. Dublin (**:05**) found the somatic nuclei of Pedicellina typically univesicular, but with two nucleoli, which were symmetrically placed in the nuclei. This, in his opinion, is not an indication of the autonomy of sperm chromosomes and egg chromosomes. He is undoubtedly justified in refusing to accept the presence of paired nucleoli as evidence of autonomy, but he seems to me to go too far in saying, that in other forms bilobed nuclei probably represent only an intermediate stage in the fusion of several vesicles into one resting nucleus. This, of course, does occur, and possibly may be true in some cases where gonomery has been claimed, but probably not in all; it is distinctly not the case in Tubularia. As already described, the daughter nuclei are double when in process of re-forming, and remain so until the next spindle is produced, and even then the chromosomes may be in two fairly separate groups.

The case in Tubularia would seem to be a good example of persistent gonomery, but what Dublin says (p. 354) in regard to Pedicellina,— viz: that in early cleavages, where autonomy should be most marked, it "is not at all in evidence" — is also true for Tubularia, and this seems to be fatal to the view of the autonomy of the sperm chromosomes and egg chromosomes in these cases. On the other hand, since in Tubularia these double nuclei occur in blastulae and in the forming germ layers when a rapid and continuous division and differentiation of cells is taking place, may it not have some connection with an intense nuclear activity? R. Hertwig (:08), referring to the "mulberry-formed" and lobed nuclei found in histology, development and pathology of metazoa, says, a comparison with similar nuclei in Protozoa shows that these nuclear forms, in the Protozoa, correspond to critical stages in the cell life, brought about by a strongly active functioning. Such an activity seems to be indicated in Tubularia at these stages, and the double nuclei may have the same relation to this cell activity. However, the symmetry, regularity and distinctness of the halves of the nuclei, and even of the definitive chromosomes, seems entirely inexplicable and superfluous on this view.

The similarity in appearance between the resting stages of double nuclei and stages in amitosis is rather close, but the history of these nuclei shows that amitosis does not occur. Divisions follow one another rapidly, and the interzonal filaments of the mitotic figure remain present for so long a time that the history of a nucleus for several generations is apparent almost at a glance. The result is that nearly all such double nuclei, as well as those apparently single, in the blastula and during the formation of the germ layers, can be proven to have been formed by mitosis, and many show an approaching, or actually beginning, subsequent mitosis. Since the same direct descent of nuclei in cleavage stages up to the blastula is nearly as certain, and, as we have seen, always leads to mitosis, we can claim with reasonable certainty, that in Tubularia crocea nuclear division occurs by mitosis from the first cleavage to the formation of the definitive ectoderm and entoderm, at least.

V. Summary.

A. PENNARIA TIARELLA.

1. Nucleolus.— The oöcyte nucleolus is a plasmatic body, which dissolves within the germinative vesicle before the nuclear membrane is ruptured. The linin network of the germinative vesicle extends to the nucleolus, so that an exchange of substances may possibly occur between the chromatin and the nucleolus.

2. Polar Cells.— Polar cells seem to be formed just before, or at

the time of, the liberation of the medusa. During the growth period the chromatin of the germinative vesicle is in fine, feebly staining granules, and a concentration of these into larger intensely staining granules and beaded strands marks the prophase of the maturation mitosis. A decrease in size and the assumption of an ovoid shape by the germinative vesicle often occurs at this time. Definite asters with centrosomes appear, the asters increase in size, and the maturation spindle begins to form before the nuclear membrane dissolves. The spindle is at first parallel to a tangent at the nearest point in the surface of the egg, and the chromosomes, in the reduced number of ten or less, are arranged in a more or less complete ring at the equator. Some of the chromosomes at least are in tetrads, which later form x- and v-shaped figures that suggest a longitudinal splitting. The spindle assumes a radial position, the asters entirely disappear, though the centrosomes remain, and a definite polar cell is detached from the egg. The chromosomes remaining in the egg now form a more or less typical resting nucleus, with membrane and nuclear reticulum, before the second spindle appears. A second maturation spindle is formed and a second polar cell is detached from the egg. The chromosomes remaining in the egg form the egg nucleus, which at first is often composed of several distinct vesicles. These may fuse, or they may remain distinct even till the time of conjugation of the germ nuclei.

3. Fertilization.— At the time of entrance of the spermatozoön the cytoplasm is very active, forming protuberances and papillae on the surface of the egg, besides definite attraction or entrance cones. The entrance of the spermatozoön, which usually occurs after both polar cells have been formed, may be at any point in the surface of the egg, but more commonly near the egg nucleus. The sperm head begins its transformation into a vesicular nucleus just within the surface of the egg, and sometimes several lobes or vesicles may be formed. The sperm nucleus in its migration toward the egg nucleus often leaves a funnel-shaped "track" in the cytoplasm. The germ nuclei are sometimes equal, sometimes unequal in size at the time of conjugation, which is by apposition. Asters and centrosomes are usually absent at the time of apposition. Polyspermy often occurs and at least two spermatozoa may form vesicular sperm nuclei. Whether more than one sperm nucleus unites with the egg nucleus could not be determined.

4. Cleavage.— The first cleavage spindle has definite polar radiations and seems to form from the cytoplasm. The second cleavage spindle is almost completely formed before the nuclear membrane is

ruptured. A large centrosphere with astral radiations is present, but no central body could be found. Cytoplasmic division is considerably delayed, the second nuclear division being finished before the first cleavage furrow has cut half through the egg.

B. Tubularia crocea.

1. Oöcytes.— The primordial germ cells divide mitotically to form the oögonia, and the latter by mitosis give rise to the oöcytes. The daughter chromosomes resulting from the last oögonial division lose their individuality, and at this time occurs a differentiation into food cells and egg cells. In the former the chromatin of the nucleus becomes scattered in large granules along a delicate linin reticulum, which is limited to the outer half of the nucleus, but is connected by linin fibres with the central nucleolus. These cells have not, perhaps, lost their power of becoming egg cells, but most of them serve as food for other oöcytes. In the oöcytes which form egg cells at once, the chromatin forms a definite spireme, which gives rise to more or less distinct loops. The loops assume a definite polar arrangement, their open ends being attached to the nuclear membrane. This apparently represents the synapsis stage, and reduction of chromosomes seems to occur at this time, though how the reduction is actually accomplished could not be determined.

a. Germinative vesicle.— The polar arrangement of the chromatin is soon lost, and the oöcyte begins to grow rapidly. The loops become granular and more delicate and may undergo a longitudinal splitting, but the evidence on this point is too scanty to allow any conclusions to be drawn. As growth continues the germinative vesicle shows no further sign of chromatin loops, and toward the end of growth exhibits only a mass of fine granules which select plasma stains.

b. Nucleolus.— The nucleolus of the oöcyte is plasmatic at all times. It increases in size for a short time in the young oöcyte (perhaps by the absorption of nuclear sap), but as soon as the oöcyte begins to grow the nucleolus may begin to decrease in size, or this decrease may not begin till a much later period. This decrease is accomplished in two ways: (1) Liquid substances pass out of the nucleolus and along the linin reticulum to become incorporated in the reticulum, presumably with the chromatin; (2) actual fragmentation may occur in the early stages, and in the later growth it always occurs. The fragments either dissolve in the nuclear sap or become arranged along the nuclear reticulum and are eventually transformed into, or absorbed by, the chromatin. The entire dissolution of the nucleolus within the germinative vesicle before the chromosomes form, suggests the possibility that the nucleolus may contain something necessary for the formation of the chromosomes.

2. Polar cells.— In anticipation of polar-cell formation the germinative vesicle at the periphery of the egg becomes reduced in size and ovoidal, and the chromatin begins to concentrate into larger masses. Two polar cells are formed, both by mitosis.

3. Cleavage.— The segmentation is total, unequal and often irregular. Polar cells, first cleavage furrow and nuclei of the first formed blastomeres occur at the same pole of the egg. The division of nuclei may be followed at once by segmentation of the egg, or nuclear proliferation may take place for some time before any cytoplasmic division occurs. These conditions are only extremes of a series and not sharply separated from each other. The cleavage may be considerably modified because it takes place within a closed gonophore.

Segmentation results in a blastula with a definite segmentation cavity, which may, however, be reduced to a few small spaces between the blastomeres, or even disappear altogether. There is a multipolar delamination of the blastula cells, and the segmentation cavity becomes filled up by a mass of cells which represent primary entoderm, the superficial cells representing primary ectoderm. The so-called morula stage in reality corresponds to the end of the formation of the germ layers, which assume their definitive positions and relations by a later specialization and rearrangement of cells.

4. Double Nuclei.— It has been possible to trace a direct descent of nuclei from the first cleavage to the germ layers, and in all cases the division is by mitosis. The nuclei of the blastula and of the germ layers are usually double, consisting of two distinct vesicles, each with a nuclear reticulum and one or more nucleoli. Chromosomes form independently but synchronously in each half and may be in two, more or less distinct, groups in the equatorial plate of the spindles. Each daughter nucleus re-forms at once into two vesicles. This condition may represent an autonomy of the sperm and egg chromosomes, but the absence of double nuclei in the cleavage stages previous to the formation of the blastula seems to be fatal to this view. Possibly this condition may be connected in some way with an intense nuclear activity.

Addendum.

Since this paper was written there has appeared a short paper by Cora Beckwith (:09) on the early history of the egg of Pennaria tiarella and Clava leptostyla. Beckwith finds that in both species polar cells are formed by a process of mitosis, which is in agreement with my results on Pennaria and Tubularia. She finds, however, that this takes place between the hours of 4 and 6 in the morning, a condition not in agreement with my observations, since I found that eggs of Pennaria which were killed at 6 A. M., and later, possessed the germinative vesicles; and only near the time of liberation of the medusa (about 7 P. M.) were maturation spindles and polar cells to be observed. This seems to show a considerable variation in the time of the completion of the maturation process.

Our results in regard to the fate of the nucleolus are also not in agreement, Beckwith finding that this body is cast into the cytoplasm, while I always found that it disappeared within the germinative vesicle before the dissolution of the nuclear membrane. Beckwith observed in Pennaria the fusion of the germ nuclei, and I, too, found this to occur, though apparently these nuclei may sometimes be separate, at the time of the formation of the first cleavage spindle. The formation of chromosomal vesicles, which often persist, and the delay of the segmentation of the cytoplasm in the cleavage of Pennaria, are observations which I can confirm.

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Explanation of Plates.

All Figures have been made with the aid of the Abbé camera lucida at a projection distance of 450 mm. The various magnifications were secured by the following combinations: × 2250, Zeiss 2 mm. apochromat. and compensating ocular 12; × 1600, Zeiss 2 mm. apochromat. and compensating ocular 8; × 51 Leitz objective 3 and ocular 4. The magnification is given for each Figure.

ABBREVIATIONS.

cl. pol, = polar cell.
o'cy^1. = oöcyte, chromatin in dense mass.
o'cy^2. = oöcyte, chromatin thread beginning; nucleolus present.
o'cy^3. = oöcyte, chromatin in a spireme.
o'go^1. = oögonia before last division.
o'go^2. = oögonia in last division.

PLATE 1.

Pennaria tiarella.

FIG. 1. Nucleus of egg from medusa killed 10 hours before time of liberation, showing large eccentric nucleolus and chromatin reticulum. Zenker, Conklin's picro-hematoxylin. × 1600.

FIG. 2. Similar to Figure 1; the nucleus ovoidal; the nucleolus almost dissolved. Bouin's fluid, Ehrlich's hematoxylin and eosin. × 1600.

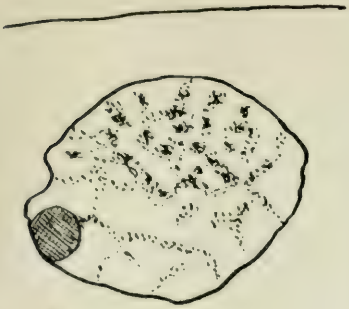
FIG. 3. Germinative vesicle of egg immediately before liberation of the medusa. Chromatin condensed into beads along the linin reticulum. Nucleolus large and homogeneous. Bouin's fluid, iron hematoxylin and Congo red. × 1600.

FIG. 4. Similar to Figure 3. Chromatin only slightly condensed, and nucleolus almost dissolved. Bouin's fluid, iron hematoxylin and Congo red. × 1600.

FIGS. 5–7. Germinative vesicles from three eggs of the same medusa before liberation, and just previous to polar-cell formation. Figures 5 and 6 possess cytoplasmic zones, but no asters. Figure 7 has two centrosomes and asters, and the formation of the first maturation spindle has begun. Bouin's fluid, picro-hematoxylin. × 1600.

FIGS. 8a, 8b. Two consecutive sections. Nearly polar views of first maturation spindle showing chromosomes in equator. Egg from same medusa as that shown in Figure 4. A splitting of some chromosomes is indicated. Bouin's fluid, iron hematoxylin and Congo red. × 2250.

FIGS. 9a, 9b. Another egg from same medusa in same condition as in Figures 8a, 8b. Some chromosomes in tetrads. Bouin's fluid, iron hematoxylin and Congo red. × 2250.



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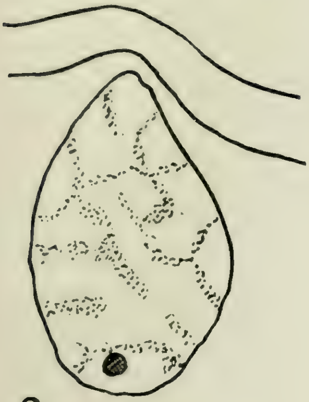

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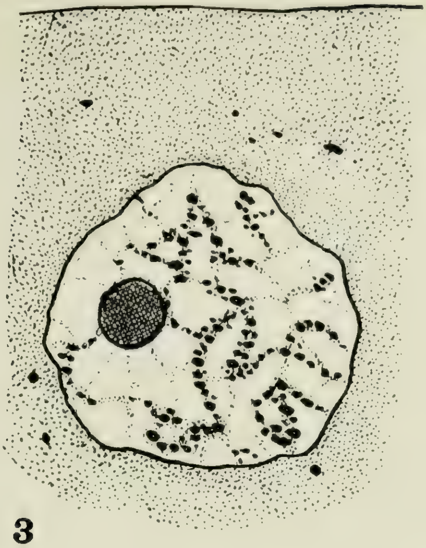

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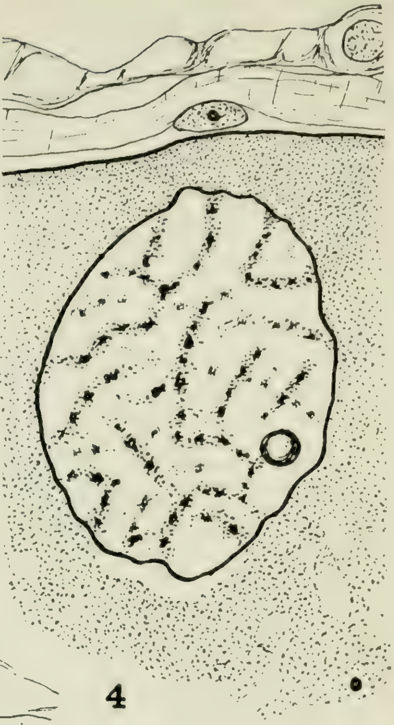

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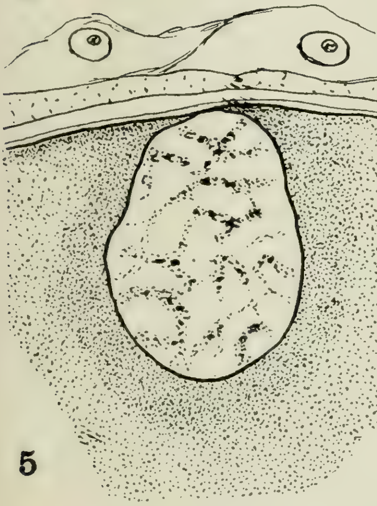

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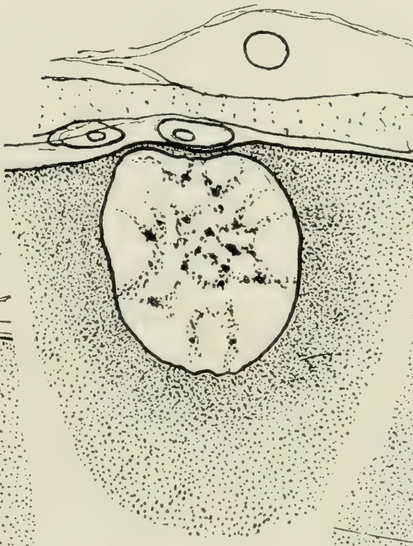

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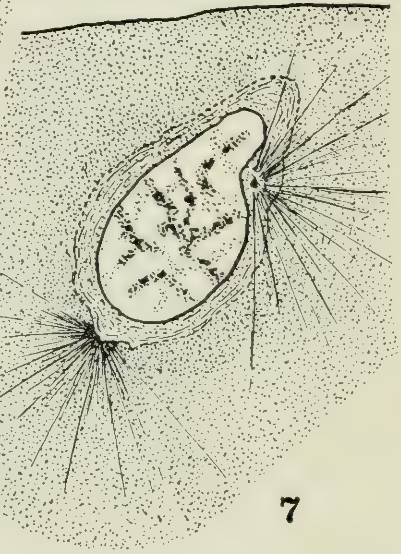

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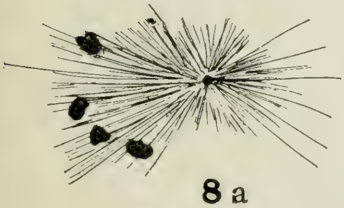

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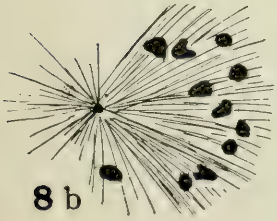

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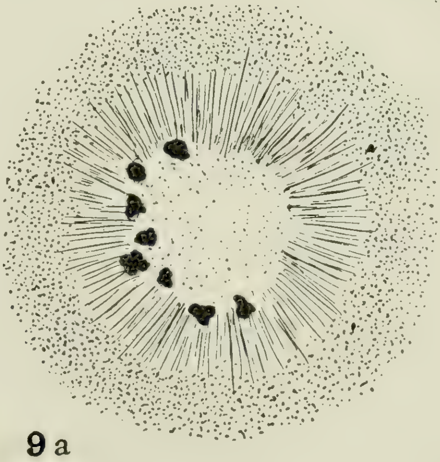

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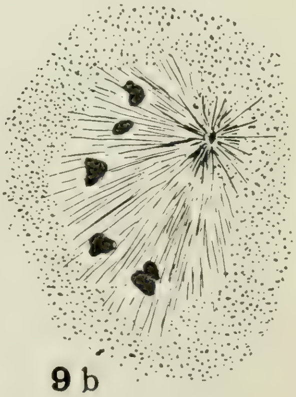


9 b

G. T. H. del.

PLATE 2.

Pennaria tiarella.

All eggs killed in Bouin's fluid, stained in iron hematoxylin and Congo red.

Figs. 10a, 10b. Egg from same medusa as that of Figures 8a, 8b. First maturation spindle. Successive sections showing chromosomes, some in tetrads. × 2250.

Fig. 11. Middle section of equatorial region of first maturation spindle. This egg had just been discharged from the medusa. Chromosomes splitting and showing x- and v-shaped figures. × 2250.

Fig. 12. First maturation spindle. Asters entirely absent; the peripheral pole shows a centrosome. Chromosomes divided and separating. × 2250.

Fig. 13. First polar cell detached; second polar cell forming. × 2250.

Fig. 14. First polar cell detached. The chromosomes in the egg form a resting nucleus before the second maturation spindle appears. Radiations are the first indications of a new spindle. × 1600.

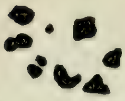
Fig. 15. Like Figure 14. The resting nucleus in several vesicles and both poles of forming second maturation spindle marked by asters. × 1600.

Figs. 16a, 16b. The two germ nuclei of one egg. Fig. 16a, the sperm nucleus with an aster and large centrosphere; Fig. 16b, egg nucleus. × 1600.

Fig 17a. Spermatozoön soon after entrance. Entrance cone on surface of the egg; a "track" behind the sperm head, and a large aster in front of it. Two other spermatozoa were present in the egg, but the cytoplasm was not differentiated about them. × 1600.

Fig. 17b. Egg nucleus of same egg. × 1600.

Fig. 18. Section of an egg showing a germinative vesicle (larger vesicle) and sperm nucleus (smaller vesicle). The only egg found where the spermatozoön was present before the polar cells had been formed. × 1600.



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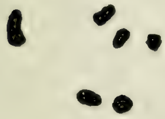

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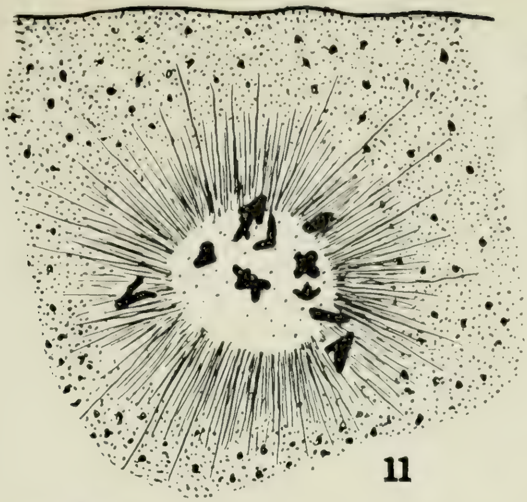

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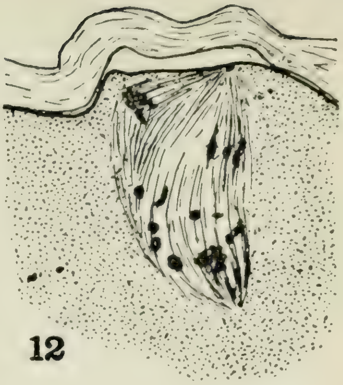

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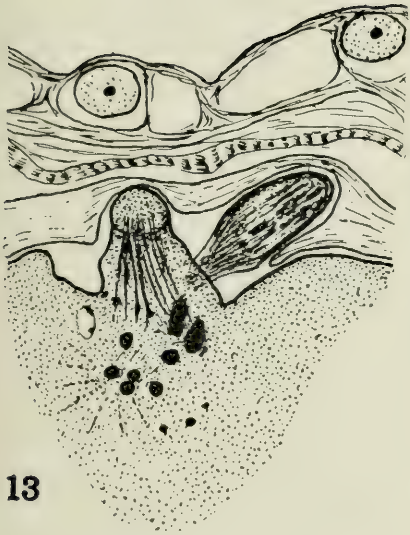

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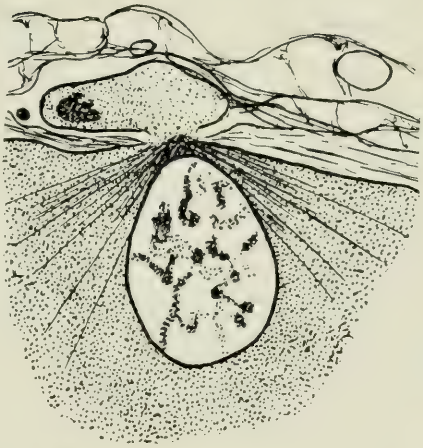

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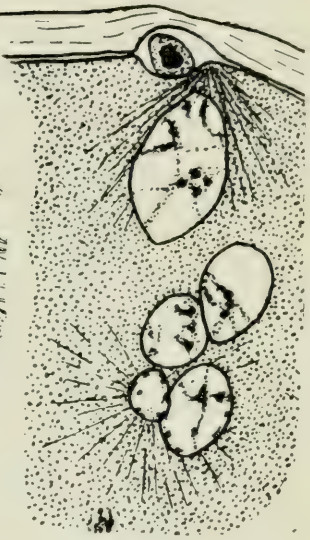

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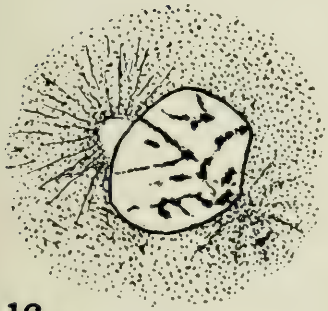

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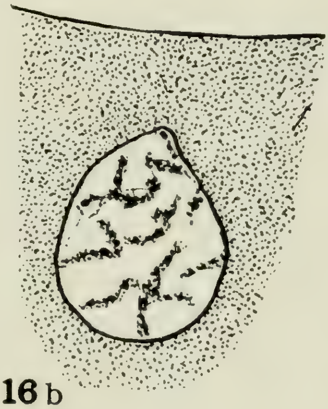

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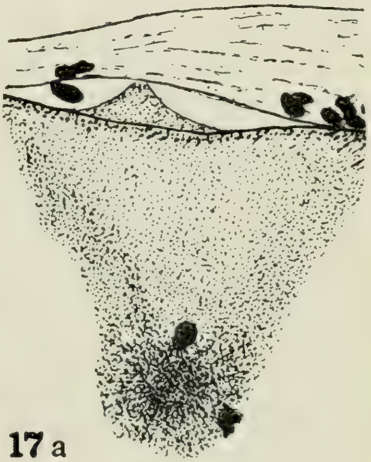

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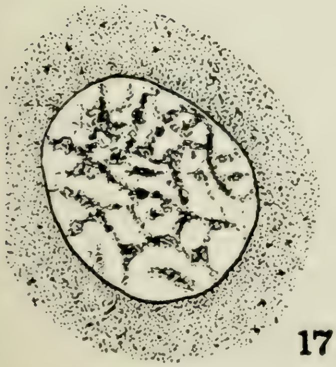

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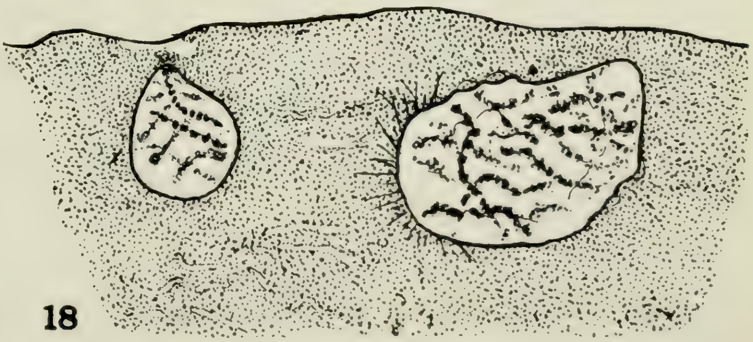


18

G. T. H. del.

PLATE 3.

Pennaria tiarella.

Eggs of Figures 19a–19c killed in Zenker's fluid, all others in Bouin's fluid. All stained in iron hematoxylin and Congo red. All except Figure 20a magnified 1600.

FIGS. 19a–19c. Three nuclei within one egg. Figure 19a shows the egg nucleus and the other Figures, the two sperm nuclei. Both sperm nuclei were near the surface of the egg, and equidistant from the egg nucleus. No asters or centrosomes accompanied any nucleus.

FIG. 20a. Portion of egg showing sperm track and three vesicles at the apex of the track. × about 190.

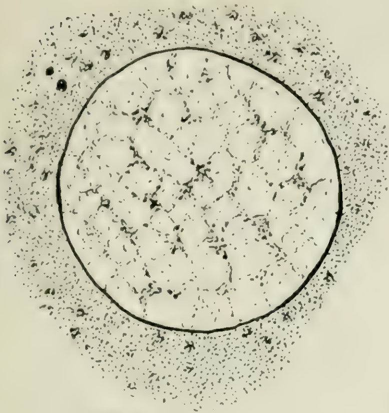
FIG. 20b. The three vesicles of Figure 20a; the middle one is perhaps the egg nucleus and the other two sperm nuclei. Although overlapping is shown in the Figure, there is no contact.

FIGS. 21a, 21b. Figure 21a is a lobed sperm nucleus without aster, and Figure 21b the compound egg nucleus of the same egg, with a large aster.

FIG. 22. Figure compiled from two sections; vesicles "a" from one section, vesicles "b" from the preceding section. These vesicles represent both sperm and egg nucleus.

FIG. 23. Germ nuclei just before contact. Chromatin in beaded strands. Neither asters nor centrosome are present.

FIG. 24. Germ nuclei of equal size, in contact. Chromatin in the form of granules in the linin reticulum. Without aster or centrosome.



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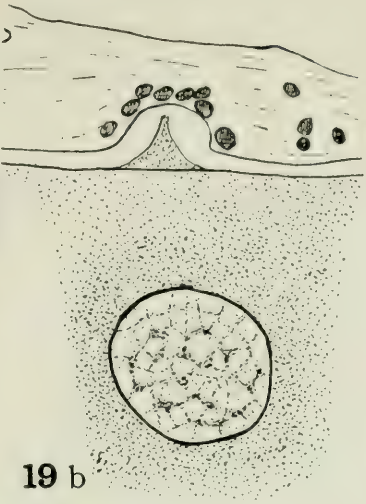

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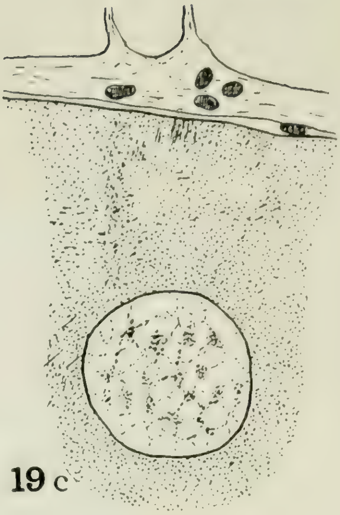

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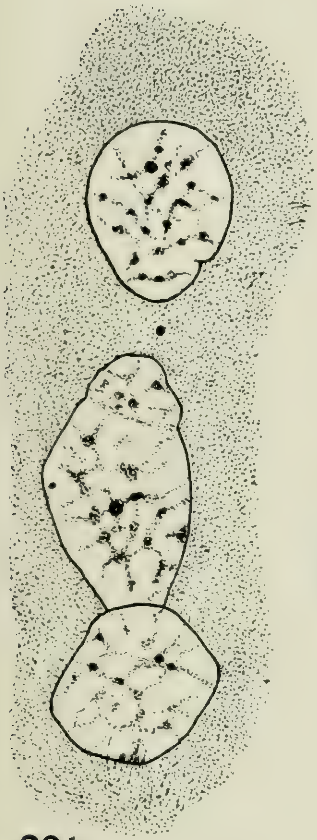

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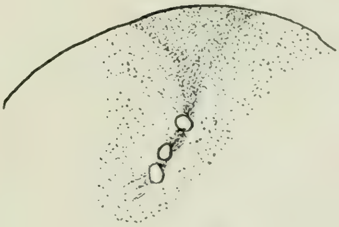

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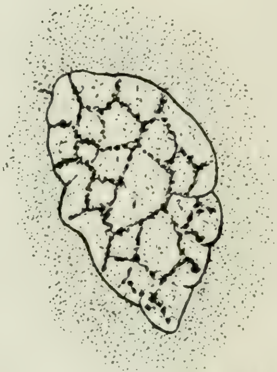

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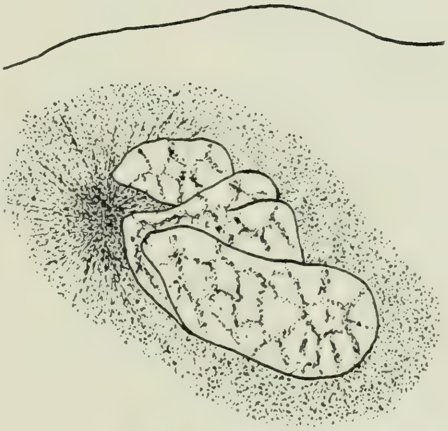

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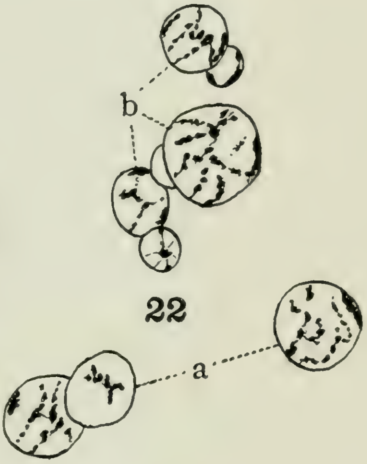

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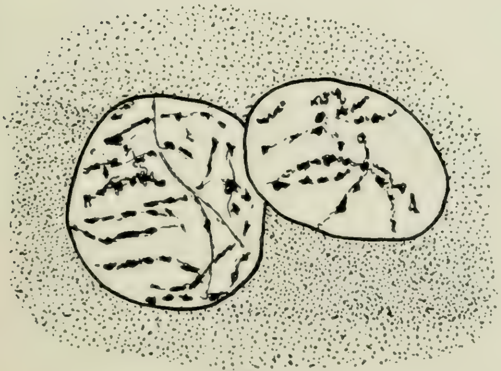

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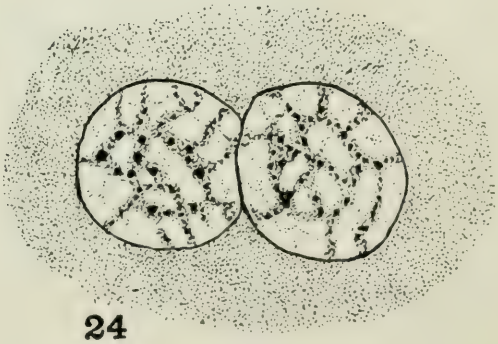

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G. T. H. del.

PLATE 4.

Figs. 25–28 *Pennaria tiarella*. Fig. 29 *Tubularia crocea*.

Fig. 25a. Germ nuclei of unequal size mutually flattened. Bouin's fluid, iron hematoxylin and Congo red. × 1600.

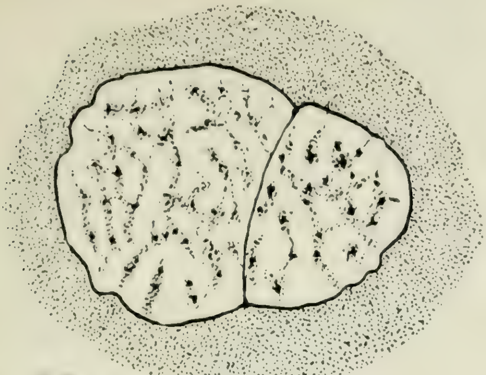
Fig. 25b. Another section of same nuclei, showing that the concentration of the chromatin begins on one side of the nuclei. × 2250.

Fig. 26. First-cleavage spindle forming. Germ nuclei have not fused; the chromatin strands, in two groups, have not yet formed definitive chromosomes. Bouin's fluid, iron hematoxylin and Congo red. × 1600.

Fig. 27. First-cleavage spindle forming. Germ nuclei have already fused into a single vesicle; chromosomes not yet formed. Bouin's fluid, picro-hematoxylin. × 1600.

Fig. 28. Second-cleavage spindle almost completed, nuclear membrane not yet ruptured. Chromosomes forming. Flemming's fluid, iron hematoxylin. × 1600.

Fig. 29. Group of germ cells from gonophore; *o'go*^{1}., oögonia before last division; *o'go*^{2}., oögonia in last division; *o'cy*^{1}., oöcyte formed, chromatin in dense mass; *o'cy*^{2}., chromatin of oöcyte beginning to form a thread, nucleolus present; *o'cy*^{3}., oöcyte after growth has started, chromatin in a spireme. Corrosive-acetic, iron hematoxylin. × 1600.



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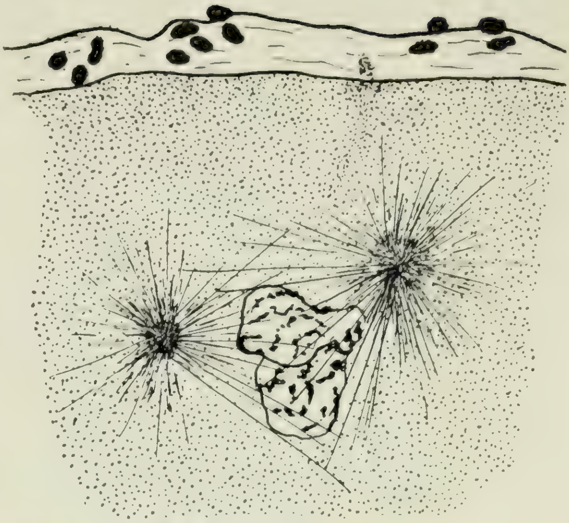

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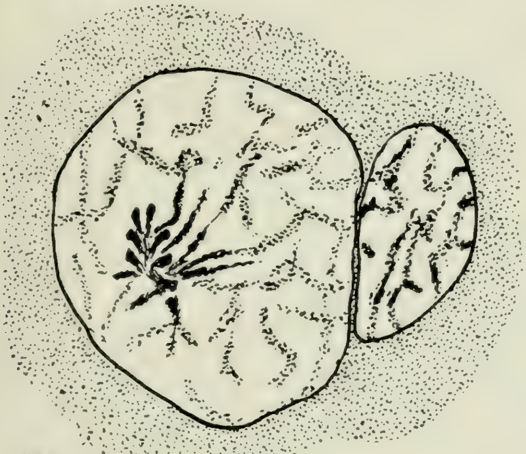

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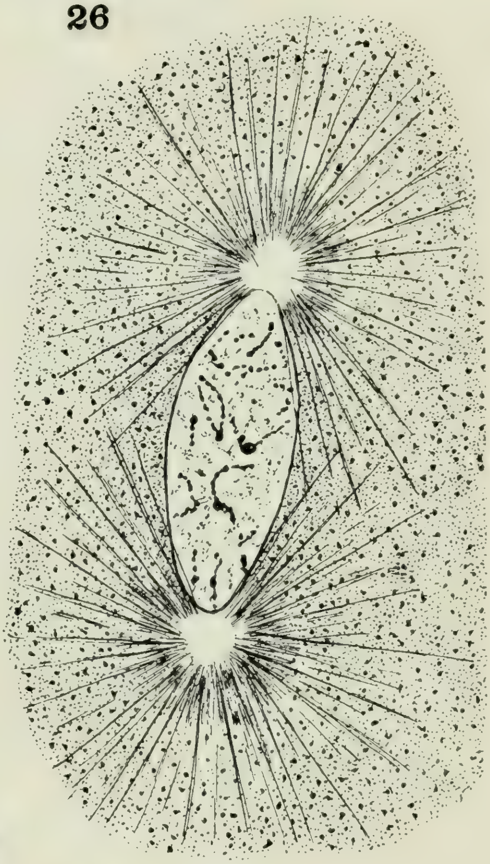

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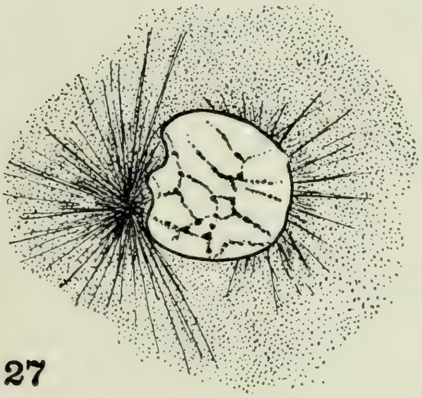

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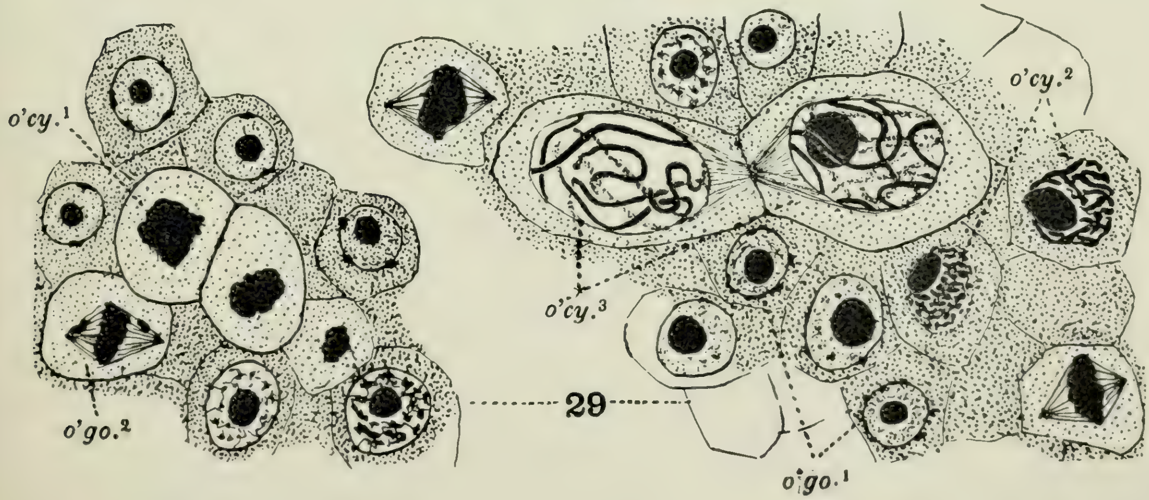


29

o'cy.¹
o'cy.²
o'cy.³
o'go.²
o'go.¹

G. T. H. del.

PLATE 5.

Tubularia crocea.

All eggs killed in corrosive-acetic. All Figures × 1600.

FIG. 30. Oöcyte before growth; chromatin has begun to form a thread. Iron hematoxylin.

FIGS. 31–33. Oöcytes in synapsis stage; loops of spireme in a polar arrangement. Iron hematoxylin.

FIG. 34. Two oöcytes; *a*, oöcyte not growing, with chromatin in scattered masses; *b*, oöcyte with loops still present but becoming granular; polar arrangement lost. Iron hematoxylin and Congo red.

FIGS. 35–37. Oöcytes at the start of the growth period. Spireme of nucleus has lost its polar arrangement. Iron hematoxylin.

FIG. 38. Oöcyte at the beginning of growth, with the chromatin in delicate granular strands. The double appearance of the threads may be accidental. Iron hematoxylin.

FIGS. 39–47. Germinative vesicles of growing oöcytes of various ages showing stages in the disappearance of the nucleolus.

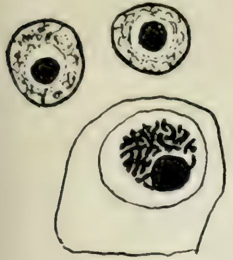
FIGS. 39, 40. Substance has left the nucleolus and is collected in several places in the nuclear reticulum. In Figure 40, *a* is the nucleus of an oöcyte which would probably have served as food; nucleolus larger than that of the growing egg, *b*. Picro-hematoxylin.

FIG. 41. Nucleolar fragments in nuclear reticulum. Ehrlich's hematoxylin and eosin.

FIG. 42. Oöcyte about one-quarter of its mature size; nucleolus in several pieces. Picro-hematoxylin.

FIG. 43. Oöcyte about half grown; nucleolus in several pieces. Ehrlich's hematoxylin and eosin.

FIG. 44. Oöcyte nearly full grown (entire egg shown in Figure 56), nucleolus in one large vacuolated, and several smaller fragments. Ehrlich's hematoxylin and eosin.



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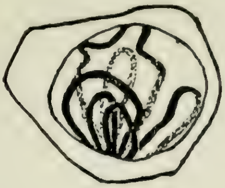

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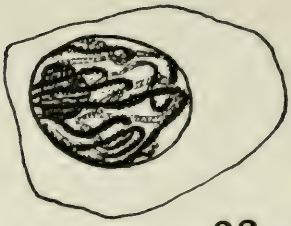

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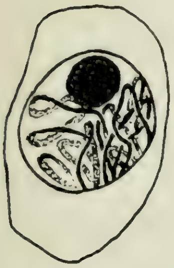

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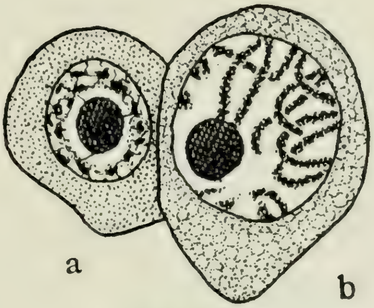

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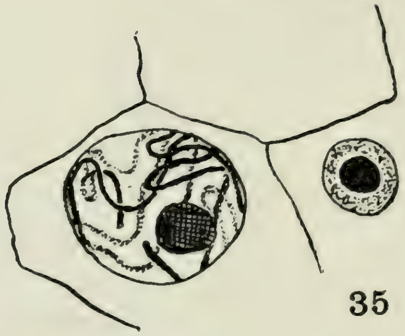

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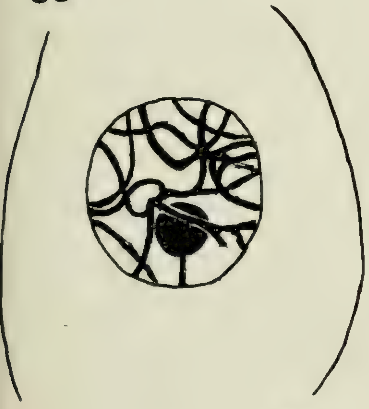

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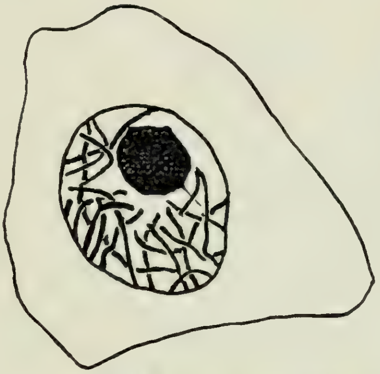

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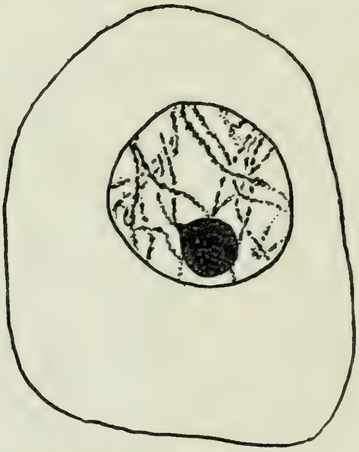

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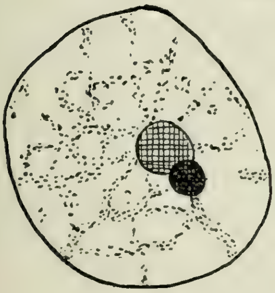

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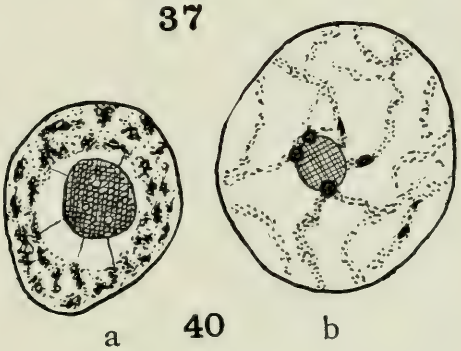

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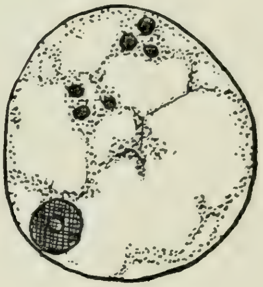

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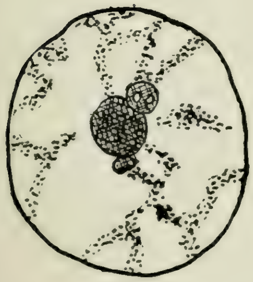

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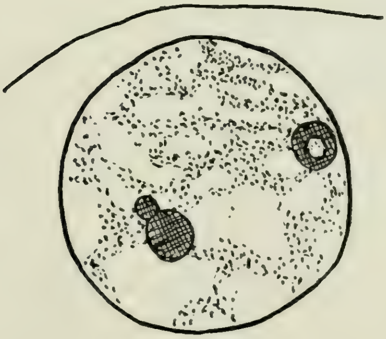

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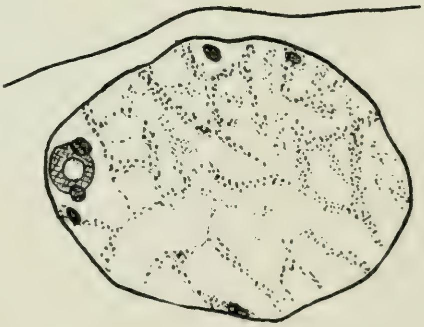


44

G. T. H. del.

PLATE 6.

Tubularia crocea.

All eggs killed in corrosive-acetic. All Figures × 1600.

FIG. 45. Egg about full grown; nucleolus of germinative vesicle fragmented into a chain of vacuolated pieces. Ehrlich's hematoxylin and eosin.

FIG. 46. Similar to Figure 45, with nucleolus in several irregular fragments. Ehrlich's hematoxylin and eosin.

FIG. 47. Extreme case of fragmentation of the nucleolus. Drawing compiled from all the sections of the germinative vesicle containing nucleoli. Ehrlich's hematoxylin and eosin.

FIG. 48. Germinative vesicle of full grown egg before polar-cell formation; chromatin in beaded strands; cytoplasm concentrated around the vesicle. Iron hematoxylin and Congo red.

FIG. 49. Germinative vesicle in prophase of maturation mitosis. The surface of the egg is raised into an elevation containing the ovoidal germinative vesicle with radiations at its peripheral end, but not directed to a visible centrosome. Cytoplasm only slightly concentrated around the vesicle. Iron hematoxylin and Congo red.

FIG. 50. Another germinative vesicle, to which the description of Figure 49 also applies.

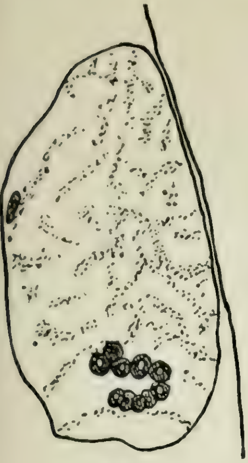
FIG. 51a. First maturation spindle; chromosomes at the equator. Iron hematoxylin and Congo red.

FIG. 51b. Sperm nucleus of the same egg as Figure 51a, no centrosome nor aster.

FIG. 52. The two polar cells already detached; the second still connected to the egg by interzonal filaments. (The polar cell shown in dotted outline occurs in the following section.) Egg nucleus near surface of the egg. Iron hematoxylin and Congo red.

FIG. 53. Somewhat oblique, nearly tangential, section. Two polar cells on the surface of the egg, and egg nucleus close to the surface. Iron hematoxylin.

FIG. 54. Sperm head with accompanying aster near the surface of the egg. Iron hematoxylin.



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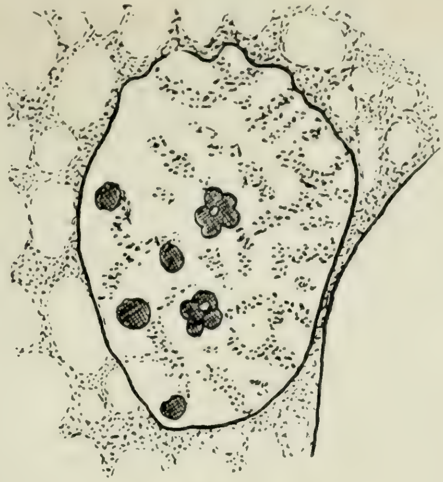

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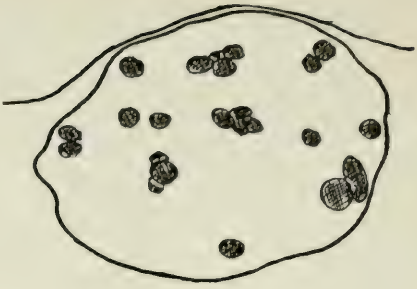

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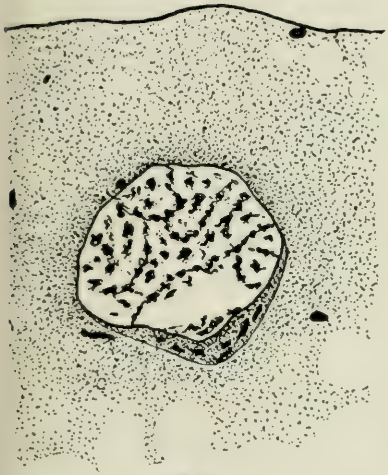

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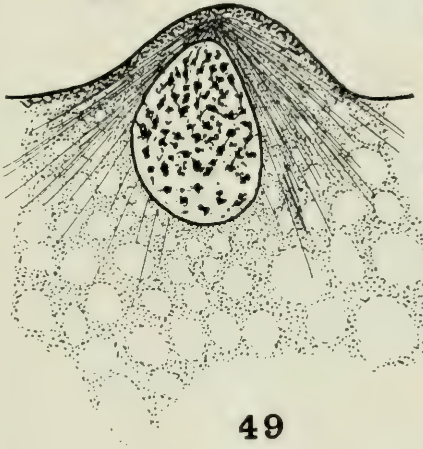

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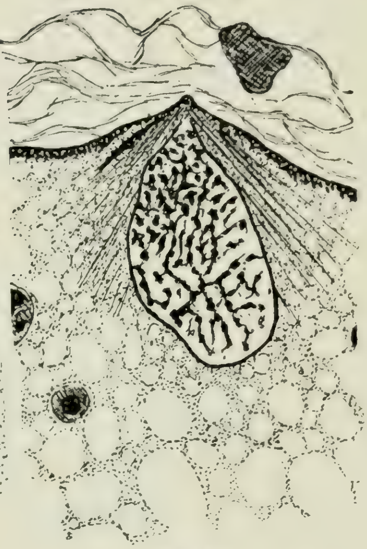

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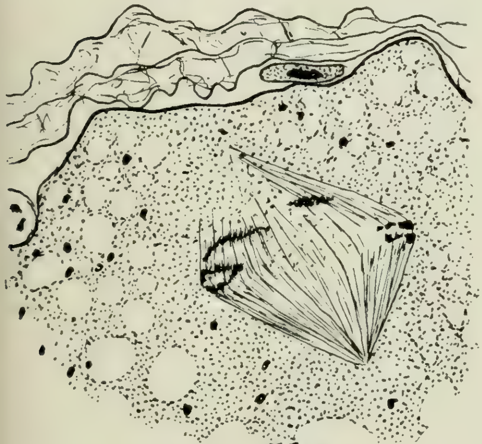

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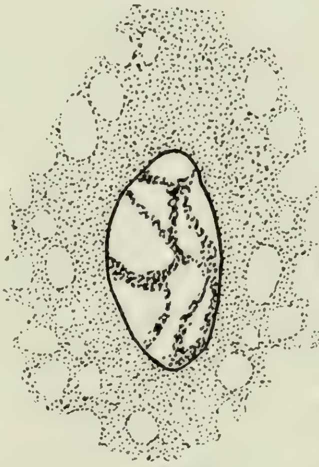

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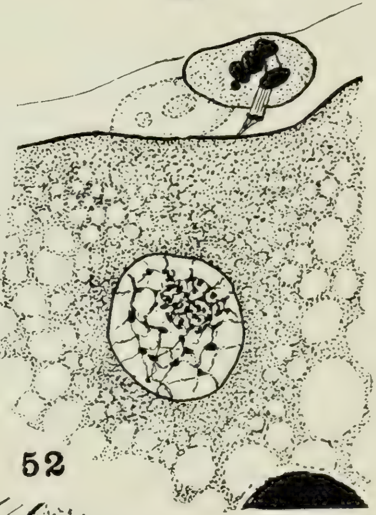

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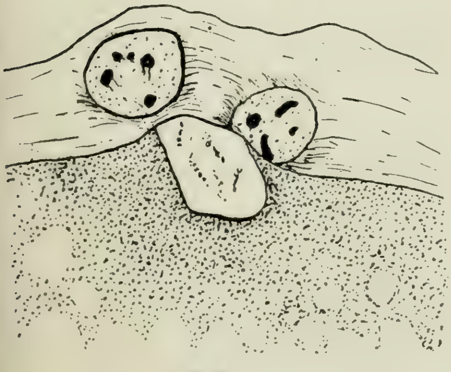

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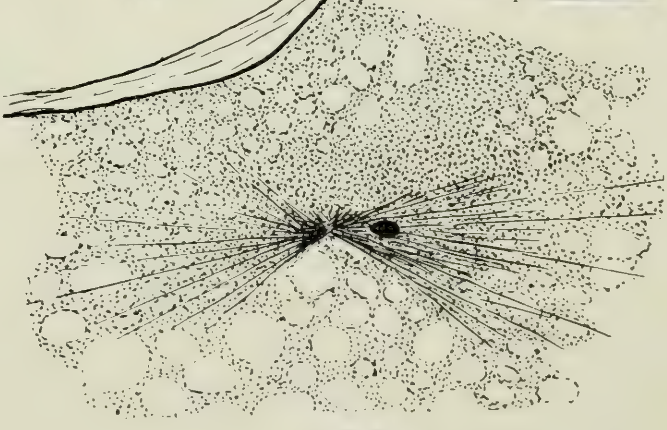


54

G. T. H. del.

PLATE 7.

Tubularia crocea.

All eggs killed in corrosive-acetic.

Fig. 55. Cross section of gonophore near the end of the growth period, showing oöcyte with pseudopodia, also spadix and other oöcytes, which serve as food. The spherical bodies in the egg are nuclei of absorbed oöcytes, "pseudo-cells." The germinative vesicle is represented by the small circle at the edge of the oöcyte opposite the spadix. Auerbach's acid fuchsin and methyl green. × 51.

Fig. 56. Like Figure 55. Ehrlich's hematoxylin and eosin. × 51.

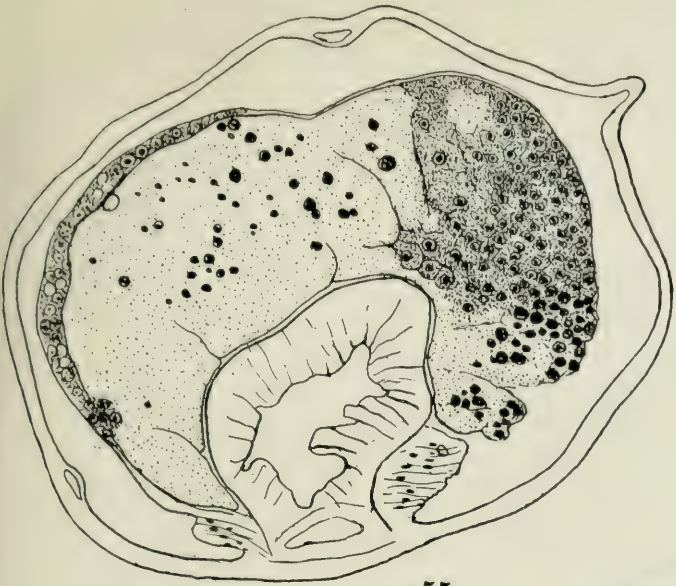
Figs. 57a, 57b. Daughter nuclei of first cleavage; each shows two vesicles. In Figure 57a one polar cell is shown. Iron hematoxylin. × 1200.

Figs. 58a, 58b. Two sections from an egg which is in an early cleavage stage. The nucleus has completed its second division. First cleavage furrow incomplete, second just started. Iron hematoxylin and Congo red. × 51.

Fig. 58c. Part of Figure 58b enlarged, showing interzonal filaments, interzonal body, one daughter nucleus, and the beginning of the second cleavage furrow. × 1200.

Fig. 59. Egg still in gonophore. Second nuclear division completed and a small blastomere forming; first cleavage furrow incomplete. In another section a similar small blastomere is found. Picro-hematoxylin. × 51.

Fig. 60. Early cleavage; 20 nuclei present, but cytoplasmic division slightly retarded. Nuclei all on the same side of the egg, and cleavage planes meridional. Cleavage cavity represented by spaces between the cells. Pseudo-cells present, but not represented in the figure. Ehrlich-Biondi mixture. × 51.



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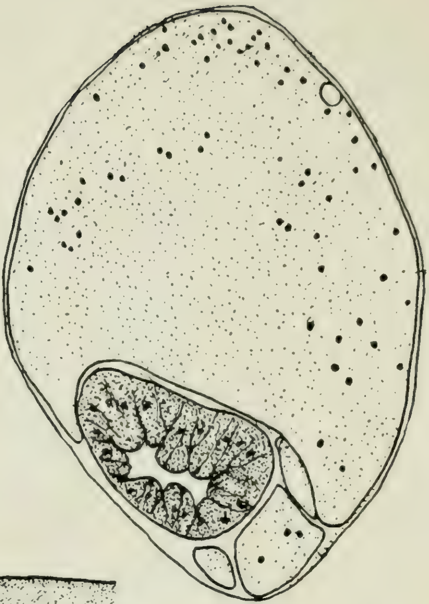

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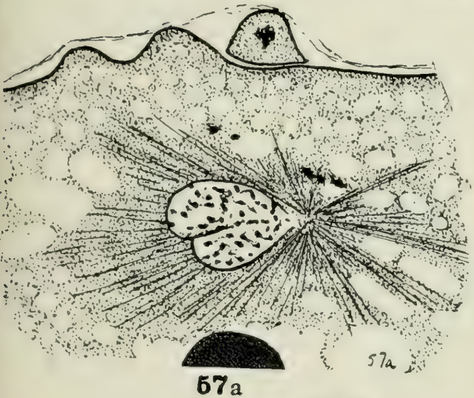

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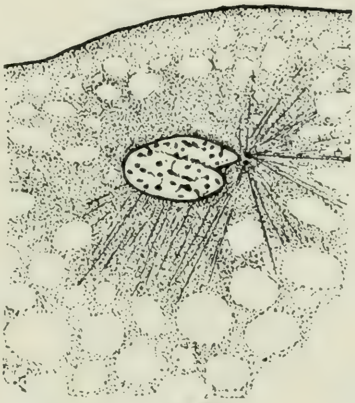
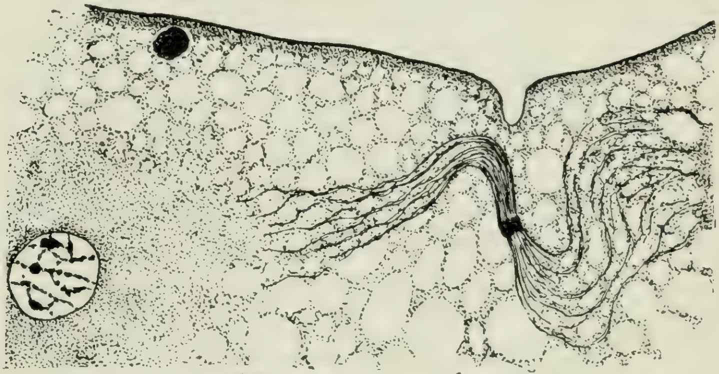


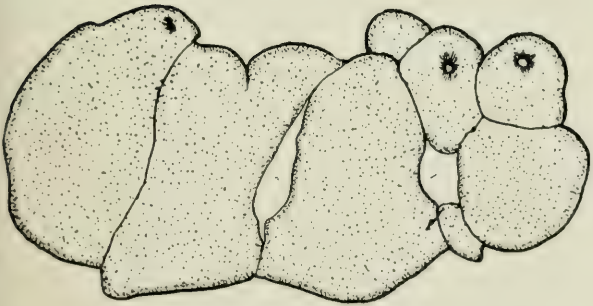
57b

58a

58b



58c



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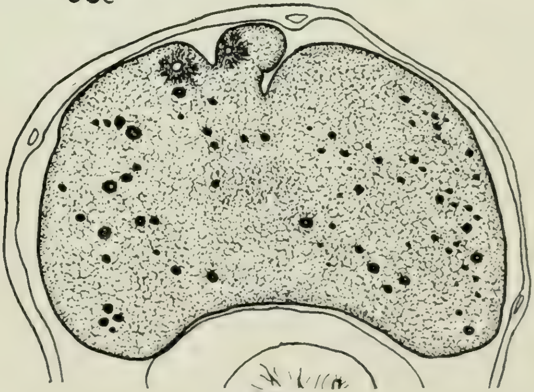


59

G. T. H. del.

PLATE 8.

Tubularia crocea.

FIG. 61. Early cleavage similar to Figure 60, but with equatorial cleavage furrows already started. Corrosive-acetic, iron hematoxylin. × 51.

FIGS. 62a, 62b. Two sections of one egg, which contained 30 nuclei, similar to Figure 61. The elongated shape of the egg is probably due to pressure in the gonophore. Pseudo-cells not represented. Corrosive-acetic, picro-hematoxylin. × 51.

FIG. 63. Blastula composed of 40–50 cells. Some of the spindles are radial, indicating the beginning of a primary delamination. The result is the formation of the germ layers and the obliteration of the segmentation cavity. Pseudo-cells still abundant, but not shown. Corrosive-acetic, iron hematoxylin. × 51.

FIGS. 64, 65. To show nuclear proliferation and the beginning of cytoplasmic division. The drawings represent the middle sections of two eggs upon which all nuclei are projected. In Figure 64 nuclei in four pairs on one side of the egg; a polar cell (*cl. pol.*) and cytoplasmic furrow occur at the same pole. In Figure 65 the nuclei are more scattered. Zenker's fluid (Fig. 64), Flemming's fluid (Fig. 65). Iron hematoxylin and Congo red. × 51.

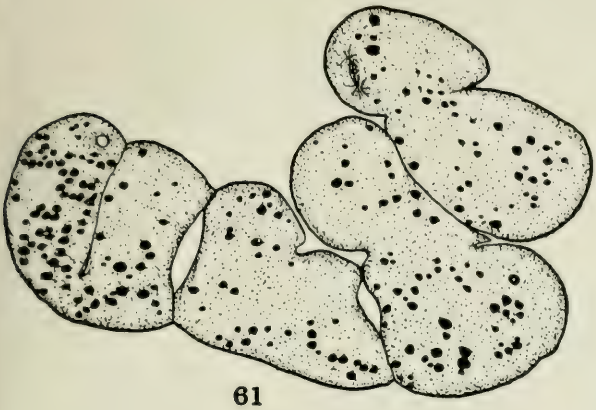
FIG. 66. An egg from the same polyp as the preceding, with a more regular, though unequal, cleavage. Zenker's fluid, iron hematoxylin and Congo red. × 51.

FIG. 67. Late stage in nuclear proliferation, cytoplasmic division in progress; a segmentation cavity present. Flemming's fluid, iron hematoxylin and Congo red. × 51.

FIG. 68. Very late stage, in which a solid mass of cells is present; cytoplasmic division still retarded. Zenker's fluid, iron hematoxylin and Congo red. × 51.

FIG. 69. More regular cleavage, in which the segmentation cavity is being filled by cells,— a part of the process of germ-layer formation. Corrosive-acetic, Ehrlich's hematoxylin and eosin. × 51.

FIG. 70. Blastula double, perhaps the result of an uncompleted first cleavage. Germ layers forming. Pseudo-cells still abundant, but not shown. Corrosive-acetic, picro-hematoxylin. × 51.



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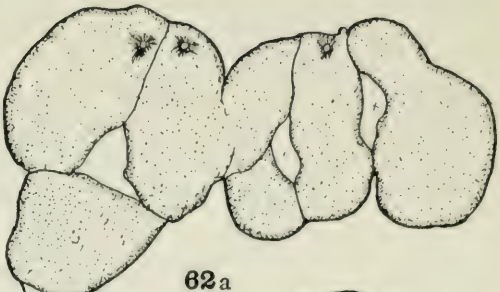

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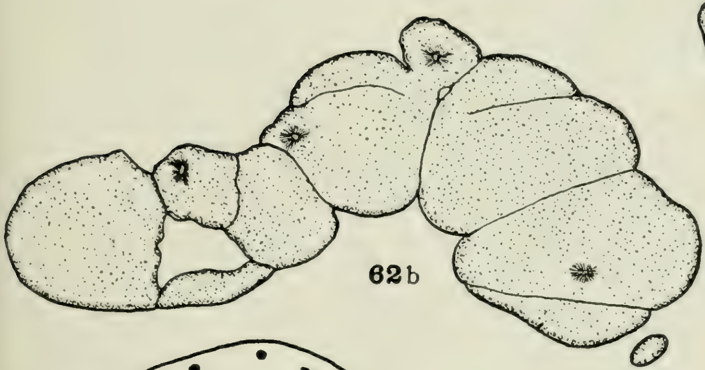

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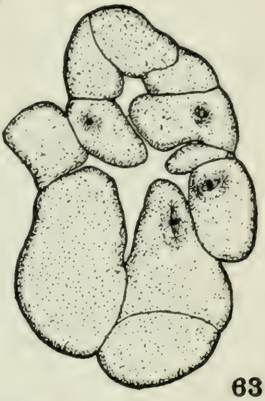

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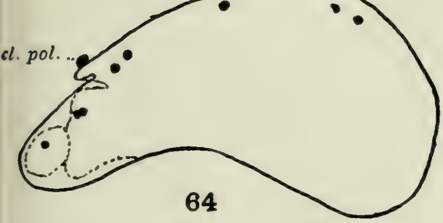

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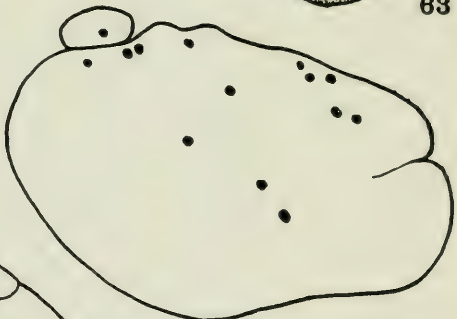

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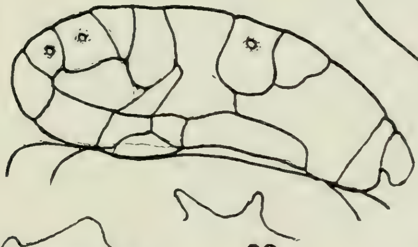

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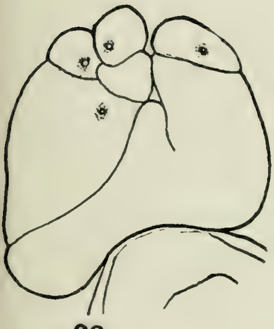

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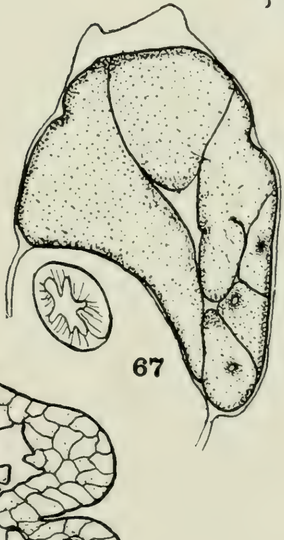

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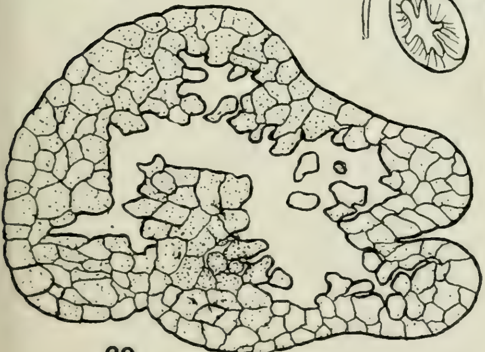

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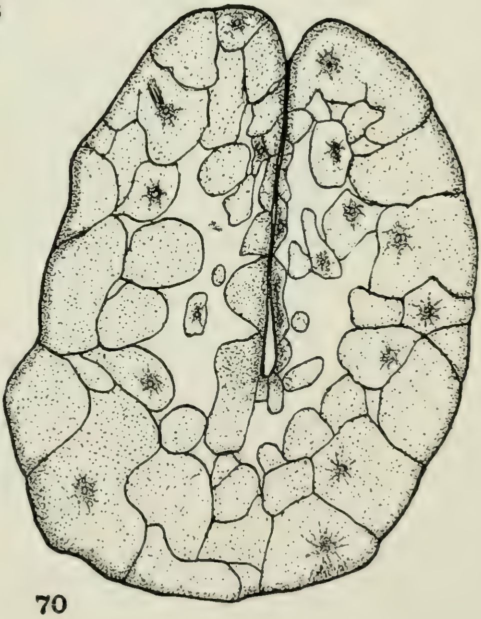

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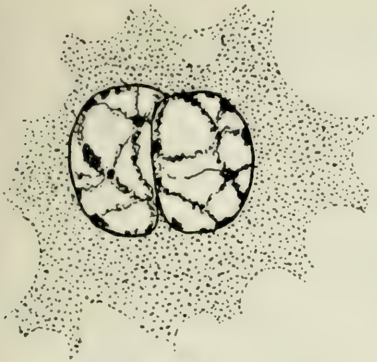
G. T. H. del.

PLATE 9.

Tubularia crocea.

All figures are from blastulae, or from embryos forming the germ layers, and show the double nuclei characteristic of those stages. Corrosive-acetic, iron hematoxylin. × 2250.

FIG. 71. Primitive entoderm cell in resting condition.
FIG. 72. Primitive ectoderm cell in resting condition.
FIG. 73. Primitive entoderm cell in resting condition, showing the persistent interzonal filaments of the preceding cell division.
FIG. 74. Entoderm cell. Spindle of an approaching division forming, and interzonal filaments of the preceding division still present.
FIG. 75. Ectoderm cell with the chromosomes formed independently in the two parts of the nucleus.
FIG. 76. Entoderm cell similar to Figure 75, but having two centrosomes and asters.
FIG. 77. Portion of an ectoderm cell showing recently completed nuclear division, interzonal filaments still visible and each daughter nucleus composed of two vesicles.
FIG. 78. Polar view of chromosomes in daughter plate, not in two groups. From a cell early in the formation of the germ layers.
FIG. 79a–79e. Polar views of five equatorial plates in the division of ectoderm and entoderm cells of the germ layers, showing arrangement of chromosomes in two groups.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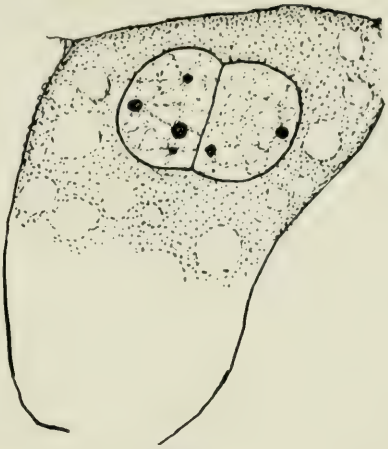
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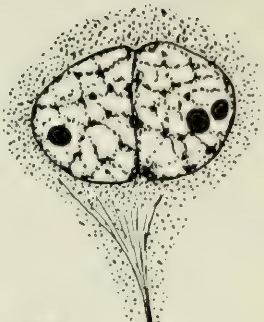
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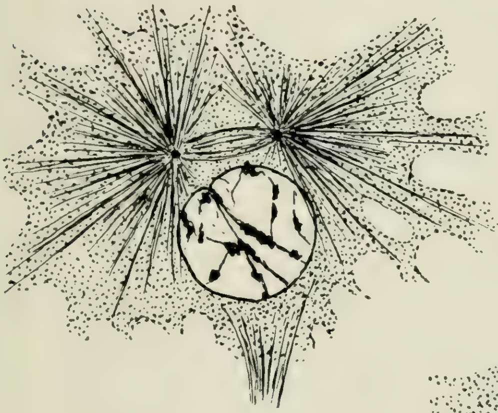
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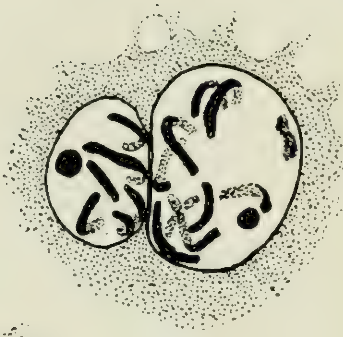
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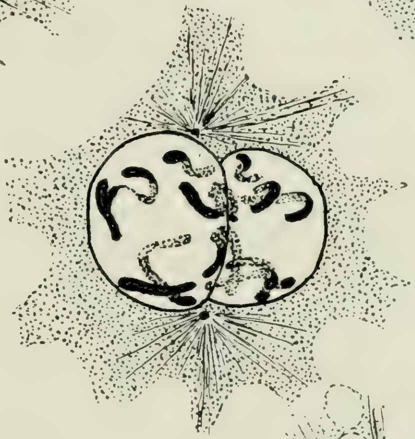
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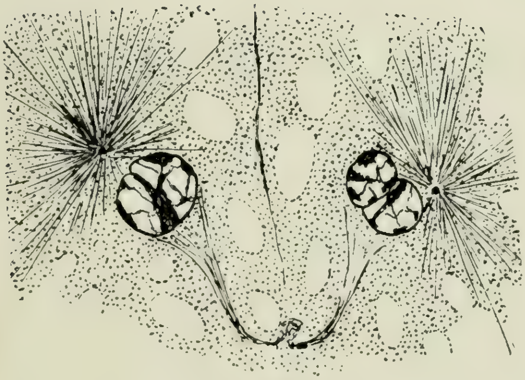
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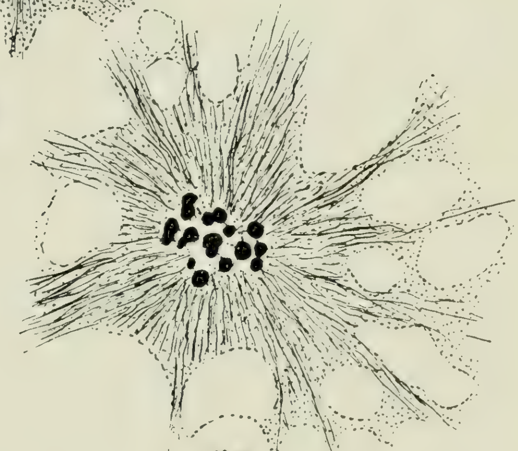
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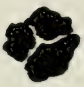
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G. T. H del.

Bulletin of the Museum of Comparative Zoölogy
AT HARVARD COLLEGE.
VOL. LIII. No. 4.

THE PERIPHERAL TERMINATIONS OF THE EIGHTH CRANIAL NERVE IN VERTEBRATES, ESPECIALLY IN FISHES.

BY R. C. MULLENIX.

WITH SIX PLATES.

CAMBRIDGE, MASS., U. S. A.:
PRINTED FOR THE MUSEUM
NOVEMBER, 1909.

No. 4.— CONTRIBUTIONS FROM THE ZOÖLOGICAL LABORATORY OF THE MUSEUM OF COMPARATIVE ZOÖLOGY AT HARVARD COLLEGE. E. L. MARK, DIRECTOR. No. 203.

The peripheral terminations of the eighth cranial nerve in vertebrates, especially in fishes.

By R. C. Mullenix.

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I. Introduction.

Since 1891 much of the investigation and discussion carried on by neurologists has been directed to the solution of the question as to the morphological units of which the nervous system is composed. The conception that the nervous system is made up of cellular elements, more or less independent of one another, which was in that year proposed by Waldeyer (**'91**) in his formulation of the neurone concept, has found wide acceptance, and stimulated investigations which have greatly advanced the knowledge of the minute anatomy of the nervous system. The theory has not escaped opposition, however, for a number of eminent men, among whom are Golgi, Apáthy (**'97**), Nissl (**:03**), and Bethe (**:04**), have vigorously attacked it; and there are not a few at present who see in the fibrillar theory a worthy rival of the neurone theory, and regard the neurofibril, and not the neurone, as affording a fundamental basis for the understanding of the nervous system.

The opposition to the neurone theory has been based largely upon investigations of the conditions in the neuropile of central organs, where Apáthy, Bethe, and others maintain that there is an intimacy of relations which is incompatible with the neurone theory. Prentiss (:**03**, :**04**) and Dogiel (:**05**) have also described conditions in peripheral nervous organs which would seem to demand the radical modification, if not the abandonment, of the neurone theory.

The present paper bears on the general problem of the validity of the neurone theory. The two cardinal questions for which I have undertaken to find answers are these: How are the terminal fibres of the eighth cranial nerve in fishes related to one another, and how are they related to the cells of the sensory epithelium in the ear sac? A condition of true anastomosis between terminal fibres of different axis cylinders would evidently be inconsistent with the neurone theory, whereas the absence of such anastomoses would tend to confirm the theory. Furthermore, a condition of organic union between sense cells and axis cylinders which are connected with cell bodies elsewhere would obviously be out of harmony with Waldeyer's conception, since he regarded the axis cylinder as an outgrowth from the ganglion cell, and a dependency of it both anatomically and physiologically. Simple contact, on the other hand, between sense cell and axis cylinder would be quite in accord with the requirements of the neurone theory.

II. Historical Review.

The relations of the terminals of the eighth cranial nerve have long been a subject of interest to histologists. The early work has been adequately reviewed by J. Kishi (:**01**), Retzius (:**05**^{a}), Kolmer (:**07**), and Bielschowsky und Brühl (:**07**), and requires only brief mention here. Among those who contributed to this early literature were Kölliker ('**54**), Max Schultze ('**58**), Waldeyer ('**72**), Meyer ('**76**), and Retzius ('**71**, '**81**^{b}, '**94**). By the use of simple stains and isolation methods these investigators were led to believe that the fibres of the eighth nerve end in the protoplasm of the hair cells, fibre and cell being continuous. The cell was regarded as the end organ of the fibre.

The application of the Golgi and the methylene blue methods to the problem resulted in a general reversal of this view. Retzius ('**92**, '**93**, '**94**), van Gehuchten ('**92**), von Kölliker ('**92**), and von Lenhossek ('**93**) came to regard the hair cells as secondary sense cells, related to the nerve fibres only by way of contact. There were still those, however, who adhered to the older view, among whom were Kaiser ('**91**), Niemak ('**92**), Ayers ('**93**), and Krause ('**96**). Ayers even went so far as to maintain that the fibres of the eighth nerve originate in the sense cells, as is the case with the fibres of the first nerve. Held (:**02**) investigated the conditions in mammals, and came to the conclusion that the sense cells are completely covered on their surface with a richly nervous neuritoplasm, which takes its origin in an intraepithelial branching of unmedullated fibres. The axoplasm is represented by him as showing a netlike structure, and as intimately grown together with the protoplasm of the sense cell.

In 1904 Ramón y Cajal (:**04**^{a}, :**04**^{c}) published a method of silver impregnation of nervous tissue which depended upon the reducing action of hydrochinon or of pyrogallic acid. Later in the same year (:**04**^{b}) he published the results of the application of this method to various nervous organs. His treatment of peripheral conditions was meagre, but a small amount of space was devoted to an account of the conditions found in the ear of chick embryos seventeen to nineteen days old. His findings were in accord, in the main, with the contact views of Retzius and others. He described two types of nerve fibres: first, giant fibres, which make their way to the summit of the cristae and expand at their ends to form a structure which is evidently identical with the "Kelchbildung" previously described by Retzius. These Ramón y Cajal regarded as separable from the sense cells with a good degree of clearness; and, secondly, fine fibres, which pass chiefly to the margin of the sensory area and end free between the peripheral ends of the cells.

Kolmer (:**04**) applied Ramón y Cajal's method to the investigation of the conditions in the ear of the frog, and obtained results not inconsistent with the contact theory. He described the sense cells as enclosed at their bases by an oval meshwork of fibrils, and, in some cases, as being surrounded at their bases or near their tops by loops of neurofibrillae. In a later paper Kolmer (:**05**^{a}) stated that upon further study, and by the close comparison of serial sections, he had concluded that the loop-shaped structure previously described was a part of a very complex pericellular network of neurofibrillae, which, however, was rarely differentiated by the method employed. He also maintained that neurofibrillae penetrate the sides of the sense cells and form intracellular networks, which, likewise, are seldom impregnated. He concluded that the fibrillae of the eighth and other sensory nerves have no real terminations, but turn back to the fibrillae of the conducting path without interruption of their continuity, by means of loops, rings, or networks. He expressed the belief that there is no distinction between the primary sense cell, as seen in the olfactory organ, and the so-called secondary sense cells of the ear, since the neurofibrillae do not lie upon the surface of the sense cells, but form a continuous network within their protoplasm.

London (:**05**) made brief mention of pericellular networks at the termination of the vestibular nerve. He cited no references to other authors in proof of such networks, but gave a single figure based upon a preparation made by the Ramón y Cajal method from the ear of the mouse. Unfortunately the figure does not represent the conditions in sufficient detail to permit of satisfactory judgment as to the real nature of the network. London concluded his discussion by expressing the opinion that it would be well to abandon the neurone theory and substitute the fibrillar theory.

Kolmer (:**05**^{b}) in a subsequent paper gave an account of further investigations, by which he sought to establish his earlier conclusions on the broader basis of comparative study. He declared that the conditions found in birds and rodents were in accord with those previously described, but not those in selachians and teleosts.

Krause (:**05**) has taken a very reactionary position upon the subject of neurological technique, maintaining that neither the Golgi method, intra-vitam staining, nor the more recent reduction methods, have brought us any nearer to a knowledge of the facts than we were twenty-five years ago as the result of the work of Retzius, with whose figures of that early time the recent ones of Krause are in close agreement. He has investigated the conditions in the ear of Petromyzon, making use of the older methods of fixation, such as the fluids of Flemming, Hermann, and Zenker, and staining with Ehrlich-Biondi's triple stain or Heidenhain's iron haematoxylin. He described the nerve fibres as expanding at the bases of the sense cells to form cups, which receive the sense cells as an egg cup receives an egg. He maintained that a mantle of nervous material surrounds the lower part of the sense cells, from which the finest kind of fibres penetrate to the interior of the sense cells. He found large and small fibres, as did Ramón y Cajal, but no evidence for the free ending of the small ones, which he described as terminating at the bases of the sense cells. Obviously Krause's conclusions are in close harmony with those of investigators who studied the problem prior to the introduction of the Golgi and other special neurological methods.

London and Pesker (:**06**) have approached the question from the embryological side in an investigation of the development of the peripheral nervous system. By a study of embryos and young of the mouse they were led to the conclusion that the fibres of the eighth nerve grow out of the cells of the spiral ganglion toward the sensory epithelium. At the same time "the impulse is given for building a fibrillar network within the sense cells, from the contained granular substance." They were led to regard the peripheral nervous system as a fibrillar mechanism which is directly united with the cerebrospinal and sympathetic ganglion cells. They were not able to satisfy themselves of the existence of free fibrillae, either intracellular or extracellular. In accord with von Lenhossék's view of the sense cells as short nerve cells without processes, they regarded the sense cells as wholly like ganglion cells, being organically united with the terminal fibres of the eighth nerve. They concluded that the results of their investigation must be regarded as furnishing further evidence against the neurone theory and in favor of the fibrillar theory.

Kolmer (:07) contributed further to the subject in a paper based on conditions found in domestic mammals. The method employed was the reduction process of Ramón y Cajal. He stated that fibrillae of a given axis cylinder are frequently in union with those of several sense cells, and that the fibrillae of a given sense cell can be shown to be in connection with fibrillae of different axis cylinders. Kolmer, unlike Ramón y Cajal, found both the giant fibres and fine fibres in the maculae, as well as in the cristae. He was not able to confirm Ramón y Cajal's distinction as to topographic distribution, inasmuch as he found both kinds of fibres in all parts of the cristae. He states (pp. 757–758) that the union between sense cell and axis cylinder appears to result from a growing together or interlacing of fibrillae which originate in the sense cell and fibrillae from the axis cylinder. A growth of fibrillae from the axis cylinder into the sense cell appears to him to be excluded, as does an outgrowing of fibrillae from sense cell to axis cylinder. He is inclined to regard the sense cells as peripheral nerve cells, and to believe that fusion between their fibrillae and those of the axis cylinder has taken place secondarily.

Finally, the most recent publication upon this subject that has come to my attention is that of Bielschowsky und Brühl (:07). The formaldehyde method of reduction of silver oxide was applied by these investigators to the ear of the guinea pig and of human embryos. In agreement with Kolmer and other investigators, they affirm that true anastomosis between neighboring fibres is not rare in the nerve plexus underlying the zone of sense cells. From this plexus some fibres pass upward between the sense cells. At their terminations they turn backward, forming a loop. A far greater number of fibres, however, are described as passing to the bases of the sense cells, where the fibrillae separate from one another and appear to clasp the cells as a bird's claw clasps a ball. This appearance, however, they believe to be due to incomplete impregnation. Cases are described in which only a few fibrillae are stained; these attach themselves to the surface of the cell, but can be followed only to the level of the upper surface of the nucleus. Besides these, they reproduce structures in which the whole cell body is enwrapped in the finest terminal twigs, which appear to be bound together by delicate anastomoses. Finally, cells are frequently found which are enclosed from base to periphery with a close-meshed trestle-work of larger and smaller fibrillae. They believe that these different appearances are due, not to the existence of different modes of termination, but rather to differences in the completeness of impregnation, and that the more numerous and more simple structures are incompletely impregnated, and that the rarer pericellular networks represent the universal terminal structure. I take it that they regard this structure as identical with the terminal expansion which has been referred to as a calyx structure. It is declared that the pericellular end structures, described by Ramón y Cajal as restricted to the summits of the cristae, have been found by them in all parts of the cristae, and are abundant in the maculae as well. Occasional sense cells are found which contain within them a definite ring-shaped body, stained like the axis cylinders. The microchemical behavior of this body leads these investigators to regard it as nervous in character. Examples were found in which this endocellular ring was connected with the pericellular network by a fibrillar bridge. The authors believe that in this structure they have found an altogether unique end-organ within the sense cell. They suggest the probability that this structure acts as a transporting mechanism which communicates the movements of the protoplasm of the sense cell to the fibrillae lying on its outer surface. The relation between sense cell and axis cylinder is not in their opinion one of contact; they prefer to describe it as one of concrescence. In the hope of being able to ascertain whether this intimate union between sense cells and axis cylinders is primary or secondary, they examined the conditions in two human embryos. One of these was obtained at the beginning and the other at the close of the second month of pregnancy. It was found that many of the primitive cells of the forming ganglion of Scarpa show processes, some of which are directed toward the periphery and some toward the brain. In consequence the investigators concluded that the union of sense cells and axis cylinders is secondarily established by the development of the extraordinary protoplasmic bridges.

III. Material and Methods.

The ear of the fish affords superior material for the investigation of this problem, in that gross anatomical conditions are less complex than in the higher vertebrates, and orientation is therefore simpler. Furthermore, owing to the absence of a bony labyrinth the terminal nervous structures are much more easy of access for chemical reagents, and complete series of sections are more easily obtainable than in other vertebrates. The fish chosen was Fundulus heteroclitus, the common "Killifish," or "Mummichog" of the Atlantic coast. The work has been done at the Zoölogical Laboratory of Harvard University, under the supervision of Professor G. H. Parker, to whom I am indebted for the inspiration, stimulating suggestiveness, and helpful criticism which characterize his teaching.

A large number of preparations was made by the older neurological methods before a process was found which differentiated the nervous from the non-nervous material of the organ. The Golgi multiple method, methylene blue, gold chloride, Vom Rath, and other methods, were resorted to, but without success. Likewise, the photographic reduction process devised by Ramón y Cajal was tried, but no impregnation was secured. Finally, resort was had to Bielschowsky's (**:02**, **:03**) formaldehyde method for the reduction of silver oxide, a method which has proved useful for the study of central organs. A large number of successful impregnations was obtained, and these preparations furnished the material represented in the accompanying drawings and constitute the basis for the conclusions arrived at.

In preparing the organ for histological treatment the head of the living fish was severed from the body, an opening was cut in the dorsal wall of the cranium to facilitate the entrance of the killing fluid, and the head thus prepared was dropped into a 12 % solution of formalin (40 % formaldehyde). Heads were allowed to remain in this fluid for at least 24 hours. Bielschowsky states that material may be preserved in it for several months, or even a year, and still give good results. An occasional renewal of the fluid is advisable.

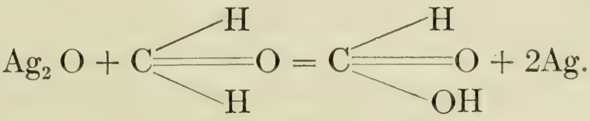
After fixation and preservation in formalin the heads were split along the median line, and each half-brain, with the corresponding ear adhering to it by the eighth nerve, was removed. By careful dissection it is possible, when desired, to remove the entire ear intact. The material was then transferred to 12 % formalin containing 1% nitric acid, for the decalcification of the otoliths, which cannot readily be removed by dissection without injury to the sensory areas. It has been found that 24 hours is an adequate time for decalcification. The carbon dioxide evolved acts as a float for the ear sac and causes it to rise to the surface of the fluid. The gas must therefore be removed, by gentle pressure, through a suitable opening made at some non-sensory portion in the wall of the ear sac. This opening also serves to admit to the interior of the ear the impregnating and reducing fluids used later, thus insuring the contact of the inner ends of the sense cells with these fluids. After decalcification it is necessary to rinse the material in flowing water for several hours, to remove all traces of nitric acid.

The material is now transferred to a 2 % solution of silver nitrate, in which it is allowed to remain for about 24 hours, after which it is removed and immersed in an ammoniacal solution of silver oxide, commonly known as Bielschowsky's fluid, for a period varying with the kind of material that is being used. This is followed by a rapid rinsing, to remove the excess of the fluid from the surface, after which the material is transferred to a 20% solution of formalin. After 12 hours or more in this fluid the material is rapidly dehydrated, cleared, imbedded in paraffin, sectioned, and mounted in balsam, in the usual way.

An essential feature of this method of silver impregnation is that the nerve paths become occupied by a finely divided precipitate of metallic silver, set free from the silver compound by the reducing action of the aldehyde. In this it differs from the Golgi method, in which there is present more or less of silver chromate, a crystalline product which is probably responsible to a large degree for the notoriously capricious results obtained by that method. It differs from the somewhat analogous reduction method devised by Ramón y Cajal (**:04**^{a}) in three rather important particulars: (1) ammoniacal solution of silver oxide is used for impregnation, in addition to the silver nitrate solution common to both methods; (2) the reducing agent employed is formaldehyde, instead of the reagents commonly used in developing photographic plates; and (3) all the processes are carried on at room temperature, rather than at the higher temperatures required by Ramón y Cajal's method.

Bielschowsky's fluid is prepared as follows: To a suitable quantity of a 2% solution of silver nitrate a few drops of a 40% solution of sodium hydroxide are added. This causes an immediate precipitate of silver hydroxide.[1] Ammonium hydroxide is now added in sufficient quantity to dissolve the precipitate of silver oxide. It is necessary to avoid more than the slightest excess of the solvent.[2] This is the solution which is commonly known as "Bielschowsky's fluid," and may be designated as ammoniacal solution of silver oxide. If it is allowed to stand for some time, a black precipitate is often deposited, which Treadwell calls detonating silver. $[AgNH_3]_2O$.

After the material has been treated with this fluid for a proper time it is rinsed quickly and then transferred to a 20% solution of formalin, as stated in a previous paragraph. In this solution, formaldehyde is oxidized to formic acid and silver oxide, in the tissue, is reduced to finely divided metallic silver, as indicated by the following equation:



$$Ag_2O + C\overset{H}{\underset{H}{=}}O = C\overset{H}{\underset{OH}{=}}O + 2Ag.$$

In order to compare the probable efficiency of the Bielschowsky method with that of Ramón y Cajal's process on chemical grounds, I have carried out the following set of simple test-tube experiments, with the results stated.

1. Silver nitrate solution (2%) was treated with a 1% solution of pyrogallic acid, at room temperature. The solution yielded after a time a slight precipitate of finely divided metallic silver. The application of gentle heat facilitated the reaction and caused a more copious precipitate.

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[1] The formation of this compound may be represented by the following equation:

$$AgNO_3 + NaOH = NaNO_3 + AgOH.$$

The silver hydroxide at once breaks down, forming water and dark brown silver oxide, as follows:

$$2AgOH = Ag_2O + H_2O.$$

In his treatise on Analytical Chemistry, Treadwell ('**03**) combines these two reactions in the following equation:

$$2AgNO_3 + 2NaOH = Ag_2O + 2NaNO_3 + H_2O.$$

[2] Dammer ('**94**) states that the chemical formula for the compound formed by this process of solution is variable, and that the product may be represented by the formula $3Ag_2O.2NH_3$ or $Ag_2O.NH_3$, or by some other formula. Treadwell ('**03**) represents the reaction by the following equation:

$$Ag_2O + 2NH_3 + H_2O = 2[AgNH_3]OH.$$

Professor H. A. Torrey, of the Harvard Chemical Laboratory, who has had the kindness to read this discussion of the chemical changes involved in silver impregnation, and who has favored me with valuable suggestions and criticisms, tells me that the fluid obtained by this process of solution behaves chemically as silver oxide in solution, and that for practical purposes it may properly be so regarded.

2. Silver nitrate was treated with a 20% solution of formalin, at room temperature. There was no evidence of chemical change. When heat was applied there was a deposit of metallic silver.

3. Ammoniacal silver oxide solution, when treated with pyrogallic acid at room temperature, yielded a heavy precipitate of finely divided silver.

4. Ammoniacal silver oxide solution, when treated with a solution of formaldehyde, immediately yielded a copious, heavy, somewhat spongy, precipitate of metallic silver, gray to black in color.

On the basis of these tests then, it appears, that there are good chemical reasons for expecting Bielschowsky's fluid to serve better than silver nitrate solution as a means of impregnating tissues with metallic silver, since it is much more readily reduced to the metallic state than is silver nitrate, whether the reducing agent be formaldehyde, or the developer used by photographers, and this at ordinary room temperature. The undesirability of leaving tissues for several days in the warm, non-preservative fluids required by Ramón y Cajal's method is obvious. While all of the reagents used in developing photographic plates are strong reducing agents, formaldehyde, is sufficiently active for the purpose, and possesses the advantage that while it is acting as a reducing agent it is also acting as a preservative.

The merit of any method, however, is determined by the results which it yields rather than by theoretical considerations. The Ramón y Cajal process has many warm advocates. I tried it without success in this investigation, but have since secured excellent impregnation of nerve terminations by its use.

My experience has been that success in impregnating the nerve terminals of the ear requires the use of a stronger solution of silver oxide and a longer treatment than is necessary for central organs. In order to arrive at once at a knowledge of the optimum conditions as to concentration and duration of treatment, I ran through a multiple series of ear sacs, varying the concentration of the Bielschowsky's fluid and the duration of treatment by regular intervals. The most satisfactory preparations proved to be those made from material which had remained for 30 to 45 minutes in a fluid prepared by adding to 20 cc. of 2% silver nitrate solution 5 drops of a 40% solution of sodium hydroxide, and dissolving the precipitate of silver oxide thus formed in the smallest possible amount of ammonium hydroxide.

Serial sections have been cut, in the usual way. The most advantageous thickness has been found to be 10 mikra. Sections of that thickness are sufficiently transparent, and permit one to study a larger part of a terminal organ at one view than is possible when the sections are thinner. This renders unnecessary the mental reconstructions from thin, serial sections, to which Kolmer (:05) was driven in his study of the conditions in the ear of the frog. It was on the basis of such a combination of a great number of sections that he arrived at his belief in the existence of an intracellular nervous network.

A further advantage possessed by sections not less than 10 mikra in thickness is that in such sections it is possible to trace axis cylinders much farther toward the brain than with thinner sections. Sections of a thickness much in excess of 10 mikra, on the other hand, present conditions so complex as to render it difficult or impossible to interpret them, owing to the great amount of nervous material which is brought into view.

By the application of a solution of the concentration already described for the period stated it has been possible to eliminate, to a great degree, the element of chance, which plays so large a part in older methods of neurological technique, and to get fairly consistent and constant results. The nervous elements are colored dark brown or black, and the surrounding tissues are usually stained a light yellow, with the exception of the nuclei and nucleoli, which, when preserved, are usually darker in color. The sense hairs are commonly well preserved, as shown in Figures 11, 12, 16 (Plate **3**), 17, 22, 26, 27 (Plate **4**), 31 (Plate **5**). There is considerable variation in the effects upon the sense cells. Like the basal cells and the basement membrane, they are usually stained light yellow, in sharp contrast to the darker color given to the nervous material. In some preparations, however, in which impregnation is good, the epithelial cells show signs of having received harsh treatment. I believe that this is due to the action of the ammonia of the Bielschowsky's fluid. The structure of the sense cells may be seriously altered, owing to the obliteration of cell boundaries, the vacuolation of the cytoplasm, and the collapse of the nuclei. Such material, while useful for tracing the courses of nerve fibres and making out the forms of nerve terminals, is not to be relied upon to any great extent in seeking to determine the relations between axis cylinder and sense cell. Figure 16 (Plate **3**) represents such a preparation. In Figure 12 (Plate **3**) a preparation is represented in which the nuclei are not well preserved and there is considerable vacuolation, but the cell boundaries are not obliterated. I have also obtained an abundance of well impregnated material in which the cell boundaries are clearly marked and the protoplasmic structures of the cell body are not essentially unlike those shown in preparations made by methods usually employed for the study of epithelial tissue. Figures 11 and 14 (Plate **3**) represent such preparations.

In many preparations which were satisfactory for determining the courses of nerve fibres and the forms of the terminal structures, it has not been possible to make out the neurofibrillae. I have obtained numerous preparations, however, in which the neurofibrillae were easily distinguishable from the axoplasm. (See Fig. 14, Plate **3**, and Figs. 21, 22, and 26, Plate **4**.)

The reasons underlying this selective activity, as it has been called, by which the silver particles become deposited chiefly in the nervous tissue, are as little understood as are those underlying the commoner methods of differential staining. It is not unusual to find it referred to microchemical changes, resulting from a special affinity between neuroplasm and silver compounds. In his discussion of the theory of staining, Mann (**:02**) states that there are some who uphold the view that the staining of animal and vegetable matter by dyes is a purely physical process, others that it is purely chemical, and still others that it is a physico-chemical process. While I am not prepared to state whether any chemical changes take place between the neuroplasm and the silver compounds or not, it appears to me that the contrast in color between nervous and non-nervous material is too pronounced to permit of being explained upon a merely physical basis, such as the possibly greater permeability of the nervous material. Preparations are not infrequently obtained in which the epithelium is so little affected that it is scarcely visible, while the nerve courses are entirely blackened with a precipitate of metallic silver. It is worthy of notice, however, that the chemical changes which result in the blackening of the nerve courses are not those which take place between the nerve and the silver oxide, if there are such. The blackening of the nerve courses is due to the reduction of the silver compound by formaldehyde. That this is true, is proved by the fact that when fresh nervous material, or nervous material which has been killed in absolute alcohol, is immersed in ammoniacal silver oxide solution there is no blackening of the tissue, whereas such tissue which has been killed and preserved in formalin is immediately changed to a dark brown when transferred to the Bielschowsky fluid. When transferred from this fluid to the 20% formalin solution, the surface of the tissue becomes still more darkened in color. If any new compound is formed as the result of chemical action between neuroplasm and the silver oxide, it is a compound which behaves toward reducing agents much as silver oxide does.

IV. General Anatomy and Histology of the Ear of Fundulus.

The anatomy of the ear of Fundulus conforms in a general way to that of Perca fluviatalis and other teleosts, as described by Retzius (**'81**^{a}) in his monograph on the vertebrate ear. The external separation between utriculus and sacculus, however, is less marked than in any species figured by him. Internally these divisions have a common cavity, excepting at the anterior and posterior extremities, where the utriculus is distinct. Anterior to the membrane which separates the two divisions posteriorly there is a horizontal shelf, which projects into the lumen of the ear, as shown in Figure 7 (Plate 2), and extends anteriorly for a considerable distance. This may be regarded as marking internally the boundary between uticulus and sacculus.

The external anatomy of the ear is shown in Figures 1–3 (Plate **1**), and sections are shown in Figures 5–10 (Plate **2**). I have followed Retzius in nomenclature, excepting that for the semicircular canals I have used the terms employed by Wiedersheim (**:07**).

The wall of the labyrinth is composed everywhere of an outer basement membrane, which is lined by an epithelial layer (Figs. 5–10, Plate **2**). In non-sensory regions this inner layer is very thin, and in many Bielschowsky preparations it can scarcely be distinguished from the basement membrane. In material stained by Mallory's (**:05**) triple stain for connective tissue, however, it is easy to distinguish the two layers; the basement membrane stains dark blue, and the epithelium takes a lighter color, is obviously cellular in character, and the nuclei of its cells are stained red.

The topographical distribution of the sensory areas and the branching of the eighth nerve for their supply agree in Fundulus with the account given by Retzius for other teleosts. In the sensory areas the epithelium is much thickened, being composed here of at least two layers of cells, the small basal cells, next to the basement membrane, and the large cylinder cells, which bear the sense hairs. Max Schultze in 1858 described the sensory epithelium in the ear of fishes as composed of basal cells and cylinder cells, with numerous nuclei between them, surrounded by protoplasm and having prolongations upwards and downwards, the former passing between the cylinder cells, and the latter between the nuclei of the basal cells. These intermediate cells he called the Fadenzellen. In subsequent publications Schultze and others described similar elements in the auditory epithelium of higher vertebrates.

In 1871 Retzius maintained that the epithelium of the maculae and cristae acusticae of fishes, amphibians, reptiles, birds, and mammals consists of only two kinds of cells,— the basal cells, which he identified with the "Fadenzellen" of Schultze, and the flask-shaped cells which bear the bristles. The former he regarded as indifferent, or non-sensory, supporting cells, and the latter as the sense cells. He represented the basal cells as bearing long threadlike processes which extend in one direction to the free surface of the epithelium and in the other direction to the basement membrane, the peripheral extensions occupying spaces between the sense cells.

Five years later Pritchard (**'76**) published an account of the histological elements of the macula acustica of the sacculus in mammals (cat), in which he described a "cuticular membrane" bordering the distal ends of the sense cells and penetrated by the sense hairs. Between the sense cells he represented triangular nucleated cells, with their bases intimately connected with the cuticular membrane and their apices prolonged downward, so that these triangular cells appeared to dovetail with the so-called "thorn cells," which were doubtless identical with what are commonly called the sense cells.

This cuticula was designated by Kaiser in 1891 as a true limiting membrane, which he described as independent of the epithelial cells.

In 1902 Held published a detailed description of the finer anatomy of the organ of Corti and other sensory structures of the mammalian labyrinth. He characterized the material found between the sense cells as supporting fibres which belong to the "Fadenzellen" of Schultze, some of the fibres extending inward to the basement membrane, others extending outward to the region of the hair cells, where they are attached to the peculiar thickened network of cuticular substance which he described as investing the sense cells.

Finally, in their recent paper Bielschowsky und Brühl (**:07**) have devoted considerable attention to the non-nervous elements of the auditory epithelium in mammals. They describe the epithelium as separated from the lumen of the ear by a limiting membrane, from which go off processes into the epithelium. These processes have the form of isosceles triangles, the bases of which rest upon the limiting membrane, while the apices reach below the middle of the hair cells to the level of the nuclei of the "Fadenzellen." Occasionally they found unstained fibres, which they regarded as belonging to the Fadenzellen. They believe these to be supporting structures, which are histochemically allied to cuticular substance. They describe the "Fadenzellen" as roundish structures with a dark, homogeneous protoplasmic body, which contains a central nucleus, with one or two nucleoli. They state that processes always extend from the body of the cell in the form of fibres, which may frequently be followed for a considerable distance toward the free surface of the epithelium, and that they sometimes contain a clear supporting fibre. These outward extending processes of the basal cells are represented as meeting the apices of the triangular projections which penetrate inward between the sense cells from the limiting membrane, and blending with them to form a homogeneous mass; a condition which leads the writers to regard these structures as a kind of intercellular substance produced by the Fadenzellen.

Inasmuch as the general histology of the ear lies somewhat outside the scope of my problem, I have devoted only such attention to it as has been indispensable to an understanding of the modes of nerve terminations. The methods which I have employed are not well adapted to the study of the non-nervous histological elements, and consequently my material does not furnish conclusive evidence concerning the matter under discussion. None of my preparations furnish any evidence for the existence of any cells in the auditory epithelium other than the basal cells and the sense cells, though it is not uncommon to find nuclei amongst the axis cylinders in the so-called inter-epithelial plexus. These nuclei, however, I believe to represent basal cells. Neither have I been able to see any processes from the basal cells extending into the spaces between the sense cells.

I have many preparations which clearly show the cuticula, or limiting membrane, separating the ends of the sense cells from the lumen of the ear. In some cases this appears to be entirely distinct from the epithelium, as in Figures 16 (Plate **3**), 18, 26 (Plate **4**) 28, 33, 36 (Plate **5**) 41, 43 (Plate **6**). In other material it is evident that there are processes extending from this cuticula into the midst of the sense cells (Figs. 38, 40, Plate **5**; 42, Plate **6**), in a manner not unlike that described by Bielschowsky und Brühl. In sections parallel to the surface these processes show well. In no case have I been able to find nuclei in this intercellular material. It often presents the appearance of being continuous with the cuticula, as shown in Figure 39 (Plate **5**). In Figure 45 (Plate **6**) material is represented in which the cytoplasm of the sense cells was much shrunken, leaving considerable space between it and the cell membranes, which remain in contact with the intercellular substance. Figure 39 (Plate **5**) represents a section cut diagonally to the principal axes of the sense cells, and hence the different layers of the macula are represented here.

Without doubt the intercellular substance under discussion is non-nervous in character. I believe that it is to a large degree composed of cuticular substance which is continuous with the limiting membrane. Whether it contains any material that belongs to the basal cells or not I cannot say.

V. Nervous Structures.

A. In Cristae acusticae.

The fibres of the ampullar ramuli of the eighth nerve, after penetrating the basement membrane, pass directly through the zone of supporting cells into what has been called the zone of plexus formation. As will be seen by reference to Figures 12, 14, 16 (Plate **3**), it is usual for the coarse, or "giant," fibres to make their way directly to the bases of the cells of the sensory epithelium, where they expand to form the "Kelchbildungen" described by Retzius. My preparations do not justify any such sharp distinction between the giant fibres and the fine fibres as was made by Ramón y Cajal (**:04**^{b}) in the case of the chick. Such extremes of size exist, but there are, as well, many intermediate sizes.

Besides those fibres which, after penetrating the basement membrane, pass directly to the bases of sense cells, there are others of different sizes, that turn in various directions to supply more remote sense cells. As a consequence there is a stratum between the supporting cells and the sensory epithelium in which the axis cylinders are so entangled with one another as to present the appearance of a real network of anastomosing fibres. This has led to the common designation of this stratum as the "zone of plexus formation." Figure 13 (Plate **3**) represents one of the most complex of such apparent networks afforded by my material. By the use of an oil immersion lens, I found, on careful focusing, that in the great majority of cases the intersecting fibres lie in different focal planes and do not anastomose. I have found some places in which conditions were not sufficiently clear to enable me to decide whether there was anastomosis or not, but I have found no place in which I was certain of anastomosis, and I am satisfied that an interlacing of fibres is the usual condition, and that anastomosis is rare, if indeed it occurs at all. I am therefore inclined to designate this region as the *zone of distribution*, rather than the zone of plexus formation.

Very commonly there is, above the zone of distribution, a stratum largely occupied by the terminal expansions of axis cylinders, designated by Retzius and other investigators as "Kelchbildungen." Such groups of terminal structures are shown in Figures 12, 14, 16 (Plate **3**). There are other places, however, in which there is no evidence of such expansions, and the sense cells appear to rest directly upon the surfaces of the fibres which constitute the so-called plexus beneath the sensory epithelium. Such a condition is represented in Figure 13 (Plate **3**).

In the cristae there are abundant terminal expansions, which are appropriately compared to the calyces of flowers, since they do not extend simply in the direction of the plane of the section, but, rather, more or less radially from the end of the axis cylinder. Consequently they are related to very many more sense cells than are visible in a single optical plane. Figure 11 (Plate **3**) represents such a structure, to which eight sense cells are visibly related. The real number of sense cells supplied by this terminal expansion is obviously many times as great as the number lying in any one optical plane. I have observed no cases of single cells being related to more than one such terminal organ.

The boundary between the sense cells and the nervous material of which the terminal expansions is composed is always distinct and sharp. There is no evidence, in my preparations, for any gradual transition from one to the other, but the dark color of the calyx ends abruptly where the yellow of the sensory cells begins. In no case have I been able to find structures corresponding in any way to the intracellular network described by Kolmer (**:05**^{a}, **:07**).

In those preparations in which the cell boundaries are best preserved it has not been possible to distinguish the cell wall in places where the cell was in contact with nervous material. Figure 24 (Plate **4**), however, represents two cells in the crista, one of which appears not to have been in contact with nervous material at its base, the cell wall being traceable with clearness to the basal part of the cell. The adjacent cell has a nerve fiber running to its base, where it bifurcates, one branch passing between the two cells, the other passing between the observer and the cell. I believe it is safe to assume that these two cells are alike in possessing a cell wall at the base, as well as at the sides, although the method employed does not enable us to distinguish that portion of the cell wall which is in contact with the nervous material, which is stained black.

Likewise, it seems probable that those sense cells which rest upon the terminal nervous expansions are separated from the expansion by cell membranes.

In Figure 14 (Plate **3**) several structures are shown in which the neurofibrillae could be distinguished. As a rule, these appear to be independent of one another, and to take more or less parallel courses. In some preparations, however, the neurofibrillae appear to form a terminal basket work. Examples of this are shown in Figures 21, 22, 26 (Plate **4**). Whether this structure is in reality a basket work, formed by the interlacing of neurofibrillae, or a structure formed by the anastomosis of neurofibrillae, is a question which I cannot now answer. I know of no way to settle such a question except by careful focusing, and in this case the diameter of the neurofibrillae is too small to permit of answering the question by that method. Such structures are rarely seen in my preparations, there being only two sections in which I have observed them. When I first noticed them I was inclined to regard them as the equivalent of the structures described by Kolmer under the name of pericellular networks. By comparing the sections in which these structures occurred, however, with the adjacent sections in the series, I came to the conclusion that they were incomplete parts of larger terminal structures, the main bodies of which lay in the next section, and that they had been obtained by cutting thin slices from the surfaces of these structures.

The terminal expansions which have been designated as "Kelchbildungen" may be regarded as a type of free termination, if I am correct in my belief that they are related to the sense cells only by contact, and that no nervous structures occur within the sense cells. They are so characteristic in their form, however, that it seems wise to class them in a category by themselves. Their expansions are chiefly horizontal, as in Figures 12, 14 (Plate **3**); but may tend toward the vertical, as in Figures 11, 16 (Plate **3**).

There is not a pronounced condition of secondary branching, such as exists in the terminal structures of the maculae. Such branches as are given off are much shorter than those of the maculae, and do not penetrate far between the sense cells. The sense cells, rather, rest upon the nervous material of the calyx, or are slightly imbedded in it.

There is another type of free termination in the cristae, which differs widely from the form just described, and corresponds more closely to what is generally understood by free terminations, in that the terminal twigs penetrate farther into the stratum of the sensory epithelium. In some cases they may be traced to fine fibres in the eighth nerve, which, after penetrating the basement membrane, make their way to their terminations between the sense cells. Examples of such fine fibres, which end free without branching, are represented in Figures

12, 15 (Plate **3**), 18, 19, 20 (Plate **4**). In other cases they originate as branches of coarse fibres (Figs. 18, 23, 25, Plate **4**). In still other cases, fibres fork at the bases of sense cells and the branches appear to clasp the bases of the cells "as a bird's claw would clasp a ball" (Figs. 15, Plate **3**; 17, 20, 23, 26, 27, Plate **4**).

In the majority of instances these terminal fibres do not penetrate far beyond the basal ends of the sense cells (Figs. 17, 19, 20, 23, 25, 27, Plate **4**). I have observed cases, however, where they extend nearly to the peripheral ends of the sense cells, or, rarely, quite to the ends. Figure 31 (Plate **5**) represents a terminal fibre which passes beyond the nucleus and middle point of the cell, on that surface of the cell which is nearest the observer. Figure 26 (Plate **4**) shows another fibre which bifurcates at the base of a cell, one branch appearing to terminate near its point of origin, and the other extending almost to the external ends of the cells between which it lies. Of course there is the possibility that the shortness of the first mentioned branch is due to the real terminal part of the twig having been severed in sectioning. In Figure 18 (Plate **4**) are shown fibres some of which reach the external cuticula, a layer often found covering the peripheral ends of the sense cells.

Whether the fibre shown in Figure 31 (Plate **5**) and the left-hand branch of the one shown in Figure 24 (Plate **4**) are superficial, or are in reality intracellular, cannot, of course, be stated with certainty for these preparations, but there is so much other evidence of the intercellular position of the nerve fibres that I feel justified in regarding these as superficial fibres which lie on the surface of the cell next the observer, and not as fibres which penetrate the cells. Figure 37 (Plate **5**) represents material which was badly shrunken by the action of the fluids used in the process of preparation, so that there are considerable spaces between the sense cells. In this case the free terminal fibres unquestionably lie in the spaces between the cells. Sections parallel to the surface of the epithelium furnish conclusive evidence in the same direction, as will be shown in a subsequent paragraph.

In most cases these intercellular branches are entirely blackened, and it is not possible to make out their fibrillar structure. Figure 26 (Plate **4**), however, shows a case in which the individual fibrillae could be seen. I do not venture to state whether the fibrillae in this case are merely intertangled, or are fused together to form a true net work. Neither do I feel justified in stating whether the individual fibrillae end free at the termination of the fibre, or turn back in a loop at the end, making a closed system of neurofibrillae, as suggested by Kolmer.

There are many places where one seems to see the free ending of neurofibrillae (Figs. 14, Plate **3**, 26, Plate **4**). Such appearances, however, might easily result from the shaving off of a portion of a terminal structure in sectioning.

B. In Maculae acusticae.

It has been decidedly more difficult to obtain material satisfactory for the determination of conditions in the sensory areas of the lagena, sacculus, and utriculus, than in the ampullae. The wall of the ear sac is considerably thinner in these regions than in the ampullae, the sense cells being considerably shorter than those of the cristae acusticae. The basement membrane is much thinner in the maculae than in the cristae and in the non-sensory areas of the ear, and is further weakened by the numerous perforations made by the penetrating nerve fibres which supply the sense cells. As a consequence of this greater delicacy of the wall of the ear sac in the region of the maculae acusticae, it turned out that in many series in which the cristae acusticae showed good impregnation the maculae were so shrivelled by the action of the ammoniacal solution of silver oxide as to render them of no value. The cases in which good impregnation has been secured and histological relations have not been badly disturbed are much rarer therefore in the maculae than in the cristae acusticae. Because of the distortion of the non-nervous tissues, there was only a single series among those in which all the nervous substance seemed to have been impregnated where it has been possible to make use of the material for the study of the maculae. I have obtained a number of series, however, the general histological conditions of which are good, in which there is an occasional impregnation of nerve fibres.

In the maculae, as in the cristae, there is, between the supporting cells and the sense cells, a stratum rich in nervous material, corresponding to the so-called zone of plexus formation in the cristae. The space between the sensory epithelium and the supporting cells is considerably less in the maculae than in the cristae, and this nerve plexus is therefore much less in thickness than in the cristae. I have not found any cases of anastomosing fibres in this zone in the maculae, but I have found many places where there was an interlacing of fibres. Cases of such entanglements of fibres are represented in Figures 29, 30, 32, 34, 35, 36 (Plate **5**).

I have not found structures in the maculae that I have been able to regard as identical with the terminal expansions (calyces) which I have described in the cristae, though giant fibres are abundant. Instead of expanding calyx-like, however, they often divide, after entering the subepithelial stratum, giving fibres which pass horizontally, and finally terminate in close relation to the sense cells. I have seen no cases in which the bases of the sense cells present the appearance of resting in concavities of these terminal structures, "as an egg rests in an egg cup." The sense cells appear rather to rest upon the surfaces of the fibres. This class of terminal structure is more suggestive of the branching of an extremely low tree than of the terminal expansion seen in the flower calyx, and may be regarded as a representative dendritic structure, or "end brush." Figures 29, 30, 32, 34 (Plate **5**) show such structures from the sacculus, and Figures 35, 36, 38 (Plate **5**) from the lagena. In some cases the fibres turn toward the sensory epithelium soon after leaving the parent fibre (Figs. 29, 30, Plate **5**). In other instances they are greatly extended in the plane of the epithelial layer (Figs. 32, 34, 35, 36, Plate **5**). As a rule the tips of these branches are turned somewhat toward the periphery, though there are cases in which this is not apparent. Often there are severed parts of such dendritic structures in close proximity to more complete ones, as in Figures 29, 30, 35 (Plate **5**).

Again, it is not uncommon to find examples of these structures in which one or more branches have been severed, leaving blunt cut ends, as in Figure 34 (Plate **5**). While cases exist where there is no evidence that the tips of the fibres penetrate between the cells, there are also many instances in which they extend for a greater or less distance into the epithelium, between the cells. There are apparent cases of the secondary branching of the fibres (Figs. 34, 36, Plate **5**).

I have found a very few instances in which there is at the termination of fibres a knob-like structure, in close proximity to the nucleus of a sense-cell. In some instances this presents the appearance of a solid mass of black, while in other cases it is possible to make out what seems to be a network of neurofibrillae. Both of these conditions are shown in Figure 36 (Plate **5**). When such a network has been observed its compactness has been such as to render its demonstration difficult. These structures have so rarely appeared in my preparations that I have not undertaken to form any opinion as to their significance.

There are other fibres of fine calibre, which penetrate the basement membrane and make their way between the supporting cells and through the subepithelial plexus to the sensory epithelium, where they end free between the sense-cells. They appear to terminate at various levels in the sensory epithelium. I have found some which could be traced to the outermost ends of the sense-cells. Such free terminations are shown in Figures 28, 33, 34, 36 (Plate **5**).

The most convincing evidence that the nerve fibres do not penetrate to the interior of the sense cells, but pass between them, is afforded by sections which lie in a plane parallel to the surface of the epithelium, or nearly so. Figures 39 (Plate **5**) and 44, 46–53 (Plate **6**) represent such sections. In Figures 44, 48 (Plate **6**) the cut ends of nerve fibres are shown. Such cut ends are always found to lie between the sense cells and not within them. Sometimes they are in close contact with the sense cells, and sometimes they are in the matrix of intercellular material by which the sense cells are surrounded. In Figures 39 (Plate **5**) and 48, 49, 53 (Plate **6**) fibres are represented which make their way between the cells of the epithelium. These conditions are typical, and are in agreement with those seen in a large number of sections. I have been able to find only two sense cells which appeared to be exceptions to the statement that the sense cells do not contain nervous material. These cells are shown in Figure 53 (Plate **6**). In one of them two fine filaments were discernible, as represented in the figure, and in the other a ring-shaped body was present, which was connected to an extracellular fibre. This latter structure at once calls to mind the intracellular rings which Bielschowsky und Brühl have figured for the sense cells of mammals. Inasmuch as I have seen only one example of such a structure, however, I am inclined to regard its occurrence as the result of come accident, and to consider it as without significance.

In the series of sections which I have just been describing, which is the single series in which the nerve fibres of the sacculus and lagena appear to be completely impregnated and the non-nervous matter is at the same time in good condition, there are many places where delicate fibres may be seen in close proximity to the surfaces of the cells (Figs. 39, Plate **5**; 46–48, 50–53, Plate **6**). Not infrequently a fibre divides at the surface of the cell, sending a branch to either side (Figs. 48, 50, 51, 53, Plate **6**). One portion of the series, including a considerable number of sections, when examined by the aid of an ordinary high power objective, gave the impression that the sense cells are completely surrounded in a meshwork of anastomosing fibres, which mark the cell boundaries. When this material was examined under the oil immersion, however, it was found that these fibres, like those of the so-called subepithelial plexus of the crista acustica shown in Figure 13 (Plate **3**), do not anastomose, but intertwine (Fig. 52, Plate **6**).

C. In Macula neglecta.

The mode of termination in the macula neglecta is represented in Figures 41–43 (Plate 6), and, as will be seen, is similar to that in the sacculus and lagena.

VI. Summary of Observations.

1. Between the supporting cells and the layer of the sensory cells is a region which is rich in nervous material in the form of an entangled mass of fibres which extend in various directions.
2. In this so-called nerve plexus I have not succeeded in finding a case in which the neurofibrillae of one axis cylinder are in undoubted continuity with those of another axis cylinder. Apparent cases of anastomosis have in almost every instance, when studied with care, proved to be due to an intertwining of fibres. The few cases which could not with certainty be so resolved may safely be regarded as due to artefact.
3. The nerve fibres which supply the maculae and cristae are of many sizes, varying from fine fibres to what have been called "giant fibres."
4. Giant fibres often expand, in the cristae, and form terminal calyx-like structures, which are associated with large numbers of sense cells.
5. Large fibres, and smaller ones as well, often branch and give rise to fine fibres, which end free amongst the sense cells.
6. It is not unusual to find fine unbranched axis cylinders passing from the place where they penetrate the basement membrane to the region of the sense cells, amongst which they end free.
7. It is not possible to make any general statement regarding the topographical distribution of giant fibres, fine fibres, and those of intermediate sizes.
8. The terminal brush of giant fibres in the maculae takes a form which is somewhat dendritic, and differs from the structure in the cristae known as the "Kelchbildung."
9. I have found no evidence of the existence of a pericellular network of nervous material.
10. I have found no evidence of the existence of an endocellular perinuclear network of nervous material.
11. The slight evidence which I have for the existence of intracellular rings of nervous substance, such as Bielschowsky und Brühl have described, may safely be regarded as due to artefacts.

12. Sections parallel to the long axis of the sense cells demonstrate the existence of free-ending axis cylinders.

13. Sections parallel, or nearly so, to the surface of the epithelium demonstrate that the nervous material is intercellular.

VII. General Discussion and Conclusions.

From the foregoing account it appears that in the ear, at least, we have a portion of the peripheral system in which the conditions are such as to furnish strong anatomical evidence in support of the neurone theory. The absence of anastomosis between different axis cylinders, the distinctness of the sense cells, and the free terminations of the axis cylinders, support the validity of that view. Though different authors have declared in a general way that anastomosis does occur, I am not aware that any investigator has maintained that the neurofibrillae of one axis cylinder are continuous with those of another, which I believe to be essential to a true condition of anastomosis. Retzius (**:05**^{b}), who has given this subject more study than any other investigator, states that he has never seen cases of true anastomosis.

The question whether free terminations exist or not depends upon what is meant by that term. Bethe and other adherents of the fibrillar theory oppose the doctrine of free nerve terminations. Dogiel (**:05**), who holds a modified form of the neurone theory, also disbelieves in their existence. The opposition is based upon the affirmed existence of peripheral networks of neurofibrillae. Thus Szymonowicz (**'96**) has described such closed terminal structures in Grandry's corpuscles. Dogiel (**:04**) has recorded similar structures in the Herbst bodies. I have already (p. 217) stated Kolmer's view, that the fibrillae of the eighth nerve have no free ends, but turn back at the periphery, forming loops, rings, or networks. If by free nerve terminations is meant the free ending of the neurofibrillae, and the maintenance of their individuality to the periphery without anastomosis with one another, my preparations do not furnish conclusive evidence in the matter. But if by free ending is meant the maintenance of the individuality of axis cylinders, or branches of axis cylinders, to the periphery without anastomosis with one another or with other elements,— such axis cylinders being composed of a greater or smaller number of neurofibrillae,— I cannot doubt that they are abundant in the region which I have studied. It is in this latter sense that the term is used by Retzius and other neuronists. Networks of neurofibrillae such as have been described by Dogiel, Szymonowicz, and Kolmer are in perfect accord with Apáthy's theory, but they are in no sense irreconcilable with the neurone theory. Likewise, the free ending of axis cylinders, while it seems to be emphasized principally by neuronists, is not at all incompatible with the fibrillar theory. The confusion in the matter seems to result chiefly from the fact of the different points of view occupied by the fibrillists and the neuronists.

The adherents of the neurone theory, upon the basis of important anatomical, embryological, and degeneration evidence, look upon the ganglion cell, dendrite, and axis cylinder as together constituting the structural unit of the nervous system. The existence of the neurofibrillae has long been recognized, but modern neuronists regard them as constituent parts of larger morphological units (Parker, :00, Collins, :06). Apáthy and his followers, on the other hand, magnify the importance of the neurofibrillae and attach less significance to the more complex structure which is known as the neurone. Apáthy represents the neurofibrillae as structures which maintain their individuality in the nerve fibre, losing their identity only in three localities, viz., in the neuropile, the ganglion cell, and the innervated organ,—sense cell, muscle fibre, or gland cell. He represents the sensory fibrillae as anastomosing in the neuropile to form an "Elementargitter," out of which larger fibrillae are assembled, which make their way to muscle fibres as motor fibrillae. From this conception Bethe draws the deduction that the neurofibrillae of the receptor organs extend without interruption to the neuropile, and from there to the motor ganglion cell, still without interruption, and are continued in the efferent nerve as motor fibrillae which make their way to the terminal organ, still without interruption. In other words, Bethe conceives that in the neurofibrillae there exists a continuous bridge between receptor and motor elements.

It is not my purpose to undertake a criticism of Apáthy's view, further than to remark that, while his conception is in some respects very plausible, there is much in it that is extremely hypothetical. To assign to the neurofibrillae, exclusively, the power of conducting nerve impulses is to make an assumption which is not supported by evidence.

I have given this résumé of Apáthy's fibrillar theory in order to prepare the way for a more critical consideration of the extracellular networks and the intracellular nervous structures which have been described by Kolmer and by Bielschowsky und Brühl. These investigators are agreed in their account of pericellular networks. In regard to endocellular nervous structures they differ.

To grant that the network which is described as surrounding the sense cells is nervous in character would not necessitate the abandonment of the neurone theory, though such a structure would of course be in perfect accord with Apáthy's conception. Kolmer admits, however, that his conclusions are based upon rare observations, and he assumes that in the great number of cells which do not show pericellular networks of neurofibrillae their non-appearance is due to imperfect impregnation. He also assumes that his inability to find evidence of such structures in fishes is due to "some specific physicochemical characteristic of fish protoplasm which renders these vertebrates unfavorable objects for the study of peripheral neurofibrillae." I know of no evidence for the truth of such an assumption. I have already pointed out good reasons why we should expect fishes to furnish the best material in the vertebrate series for the study of this problem.

Bielschowsky und Brühl, like Kolmer, base their belief in the existence of pericellular networks upon rare observations, assuming that the more numerous and more simple structures are the result of imperfect impregnation, and that the rarer pericellular networks represent a universal terminal structure. This appears to me to be a dangerous assumption, and I believe that the safer course would be to look with suspicion upon structures of such rare occurrence, regarding them as probably artefacts. It would perhaps appear presumptuous for me to venture a criticism upon the handling of a method which Bielschowsky himself worked out. Nevertheless, it should be recognized that, superior as this method is to the older impregnation processes, it is easy, by overdoing the treatment with the silver oxide solution, to cause almost any tissue to take on the color characteristic of nervous matter in good Bielschowsky preparations. I have several times obtained preparations in which the sense hairs, and the sense cells as well, were entirely blackened, and have seen preparations from the lateral line region in which muscle fibres were well impregnated, the cross striations being clearly differentiated. I have no doubt that by excessive treatment with silver oxide solution it would be entirely possible to cause non-nervous intercellular material to take on the appearance of nervous substance. What constitutes successful impregnation is a matter which it is necessary for each investigator to determine for himself, and it is this last resort to the personal judgment of the investigator which is responsible for many of the differences of opinion in this field. The controversy between the neuronists and the fibrillists has resolved itself largely into the question whether certain structures described by Prentiss, Bethe, and others, are in reality nervous in character. It is not uncommon to find adherents of the neurone theory maintaining that the fibrillists have been misled by over-impregnation, and it is no less common to find adherents of the fibrillar theory attributing the conclusions of neuronists to the incomplete impregnation of their material.

It may well be recognized that the characteristics of nervous material by which it may be differentiated from non-nervous material, in the study of neuro-histological problems, are not so numerous nor so varied as might be desired. A good deal of what we believe concerning the physiology of the sense cells and nerve fibres, particularly in the ear, rests upon analogical evidence which has not as yet been confirmed by experiment. It is probably safe to assume, however, that both the sense cells and the axis cylinders possess the physiological properties of nerve tissue, which Gotch, in his article in Schäffer's Physiology (:00), has characterized as "that tissue which exhibits phenomena of excitability, conductivity, and states of excitation." Without doubt these characteristics are possessed in common by sense cells and nerve fibres, though probably in varying degrees. Since it is as yet impossible to obtain direct evidence as to the functional differentiation between them, we are forced to content ourselves with such morphological evidence as is afforded by differential staining, and to assume that those portions which exhibit peculiar affinity for silver compounds are nervous in character, as is undoubtedly the case in other places in the animal body, and that those which do not exhibit such affinity are non-nervous. Such a distinction is of course entirely inadequate as a general definition of nervous material, but it constitutes the best evidence that is obtainable at present, and it has seemed to me that it is well to recognize the narrowness of the tests upon which we are forced to rely in attempting to solve problems of this kind.

My conclusion, then, regarding pericellular networks, is that the nervous character of such networks is doubtful, but that no violence would be done to the neurone theory by their confirmation.

The theoretical significance of intracellular networks of neurofibrillae, such as Kolmer has described, and of such intracellular structures as Bielschowsky und Brühl have described, would depend to a large degree upon their embryological origin and the relations of the neurofibrillae of the sense cells to those of the axis cylinders. If it should be shown that the sense cells of the auditory epithelium are not secondary, but primary, sense cells, as is the case in the olfactory epithelium, it would seem that Apáthy is right in ignoring the cell bodies and regarding the neurofibrillae as the structures of primary importance, for then we should have two cell bodies corresponding to a single axis cylinder, and there would be no reason for regarding the neurofibrillae as dependent upon either cell body in any peculiar way. On the basis of such embryological evidence as has thus far been adduced, however, it appears that such relation as exists between the alleged neurofibrillae of the sense cells and those of the axis cylinders is secondarily established. The embryological investigations of London und Pesker (:06) to which I have already referred (pp. 218) have led them to view the alleged neurofibrillae of the sense cells as originating within the sense cells, and those of the axis cylinders as coming from the ganglion cells. They found no evidence for the existence of fibrillae except in association with cell bodies,— either ganglion cells or sense cells, which latter they regarded as wholly like ganglion cells.

Likewise Kolmer, as I have stated on page 219, affirms that the union between sense cell and axis cylinder appears to result from a growing together or interlacing of fibrillae some of which originate in the sense cell while others come from the axis cylinder. He believes that the union between the fibrillae of the sense cells and those of the axis cylinder is established secondarily, and is not the result of the growth of fibrillae from the axis cylinders into the sense cells or from the sense cells into the axis cylinders. He regards the sense cells as peripheral nerve cells.

Finally, the embryological evidence obtained by Bielschowsky und Brühl bears out the conclusion that such union as exists between axis cylinders and sense cells is secondarily established.

In view of this embryological evidence I am not able to understand why Kolmer, and London und Pesker consider it necessary to abandon the neurone theory, even if all the peripheral networks which have been described for this organ should meet with confirmation. They themselves regard the sense cells as peripheral nerve cells, which view is in complete accord with the interpretation given them by von Lenhossék, who has regarded them as short neurones.

However, I am not yet prepared to accept these intracellular structures as established. The testimony to their existence is scanty and inharmonious, the structure which Kolmer has described being totally different from those described by Bielschowsky und Brühl, who refuse to accept Kolmer's results, characterizing them as an admittedly chance product, and suggesting that the appearance of the structure which Kolmer has interpreted as an intracellular network of neurofibrillae may be due to over heating in the warm solution of silver nitrate which is used in the Ramón y Cajal process.

The intracellular rings and the protoplasmic bridges connecting them with the axis cylinders which Bielschowsky und Brühl have described are unique, having been recorded by no other investigators. The observation is interesting, in that it affords a suggestion of histological evidence in support of the theory of the synapse between neurones, which has been advocated by Sherrington (:06). It has been found that nerve conduction in which two or more neurones are involved is much slower than conduction along an axis cylinder of a single neurone. Mislawsky ('95) found that the direction of the nervous impulse is reversible in nerve-trunk conduction, whereas in reflex-arc conduction it is irreversible. These and other physiological differences between nerve-trunk conduction and reflex-arc conduction Sherrington has referred to that part of the arc which lies in the central gray, and he has introduced the term synapse to represent those "intercellular barriers" in the central gray which constitute the nexus between neurone and neurone. If the view that the sense cells are nervous in character should prove correct, the protoplasmic bridges which Bielschowsky und Brühl have described in the periphery might be regarded as representing the synapse for which Sherrington adduces so much physiological evidence in the spinal cord. The neurones involved in audition are so short and other conditions are such as to render it impracticable to obtain, in this structure, physiological evidence of the kind that Sherrington has obtained by experimentation upon spinal nerves. But the confirmation of the existence of the protoplasmic bridge would be interesting because it would supplement the physiological evidence obtained from other regions.

I must say, however, that I do not believe that the existence of either the intracellular rings or the protoplasmic bridges can be regarded as established until confirmed by other investigators. My preparations furnish no evidence of the existence of the protoplasmic bridges, and only slight evidence of intracellular nervous material.

In conclusion, it may be stated that in so far as Bielschowsky preparations may be relied upon as revealing the actual conditions, we are justified, in the case of the fishes, in regarding the relation between nerve terminals and sense cells as one of contact, and not of organic union, and the relation between different axis cylinders as, likewise, one of contact rather than continuity. Such preparations make it possible to trace nerve courses more completely than do Golgi preparations, and there is the additional advantage that neurofibrillae are differentiated, as they are by methylene blue. My results bear out the conclusions of those investigators who have studied the problem by the Golgi method, and do not lend support either to the view of pericellular networks of neurofibrillae, nor of intracellular neurofibrillar structures.

In stating these conclusions I do not wish to be understood as holding that sense cells are morphologically or physiologically independent of axis cylinders. It is now recognized that even in those structures in which there seems to be the greatest distinctness of cell limits, the cells are far less independent of each other than was believed in the earlier days of the cell theory. So far as it is safe to judge from the conditions found in the ear, however, I believe that we may say that the cell theory is as applicable to nervous tissue as it is to other tissues of the animal body, and that the neurones, which are to be thought of as the peculiar and complex cells of which nervous tissue is composed, are as independent of one another, and as independent of other kinds of cells, as are the cells which compose many other animal tissues.

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Explanation of Plates.

All drawings were made with the aid of the camera lucida.

The magnification for Plate 1 was obtained with a Leitz dissecting microscope; for Plate 2, a Reichert objective No. 3 and ocular No. 4 were used; and for Plates 3, 4, 5, and 6, a Reichert $\frac{1}{12}$ inch homogeneous immersion objective and ocular No. 4.

ABBREVIATIONS.

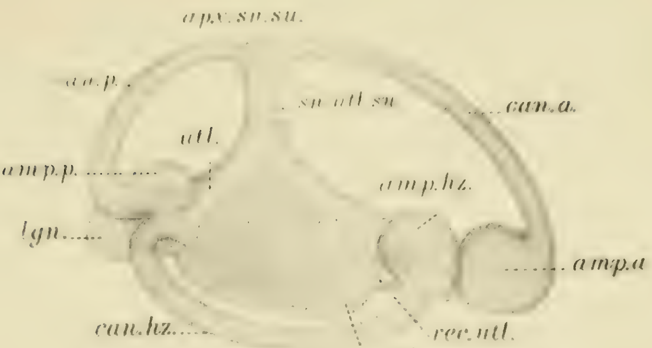
amp. a.	Ampulla anterior.
amp. hz.	Ampulla horizontalis.
amp. p.	Ampulla posterior.
apx. sn. su.	Apex sinus superioris.
can. a.	Canalis anterior.
can. hz.	Canalis horizontalis.
can. p.	Canalis posterior.
cr. ac.	Crista acustica.
lgn.	Lagena.
mac. lgn.	Macula acustica lagenae.
mac. ngl.	Macula neglecta.
mac. rec. utl.	Macula acustica rec. utriculi.
mac. sacl.	Macula acustica sacculi.
n. oct.	Nervus octavus.
rec. utl.	Recessus utriculi.
rm. a.	Ramus anterior.
rml. amp. a.	Ramulus amp. anterioris.
rml. amp. hz	Ramulus amp. horizontalis.
rml. amp. p.	Ramulus amp. posterioris.
rml. lgn.	Ramulus lagenae.
rm. p.	Ramus posterior.
sacl.	Sacculus.
sn. utl. su.	Sinus utriculi superioris.
utl.	Utriculus.

PLATE 1.

Magnification, 10 diameters.

External appearance of right ear of Fundulus.

Fig. 1. Lateral view.
Fig. 2. Dorsal view.
Fig. 3. Ventral view.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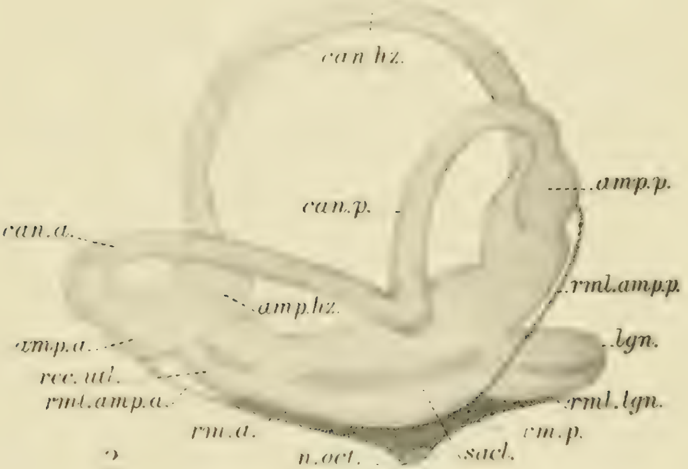
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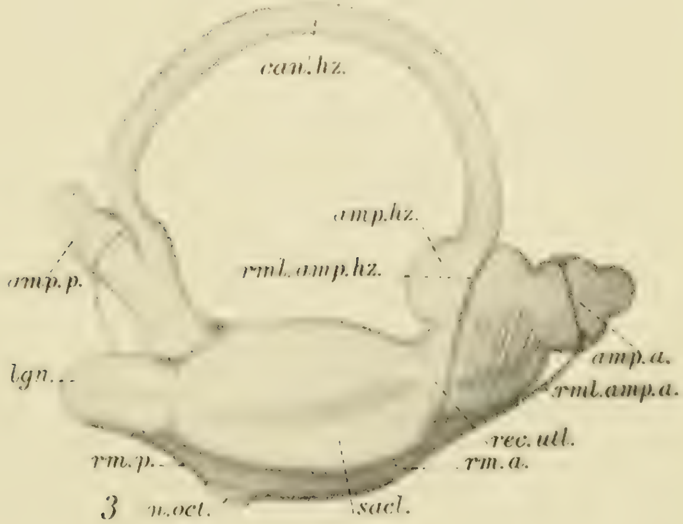
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sacl.

3

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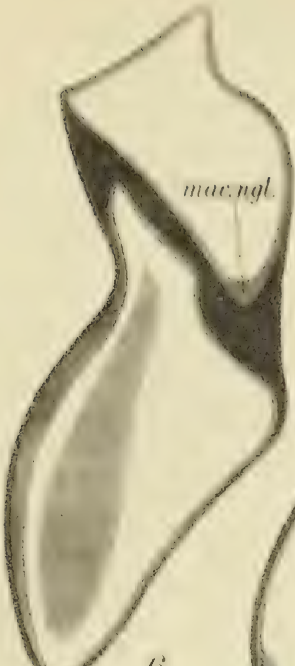
PLATE 2.

Magnification, 60 diameters.

Series of sections through the ear sac. The basement membrane is represented by the darker tint. The inner, lighter, tint represents the epithelial lining of the membranous labyrinth, which is non-sensory in the thinner regions and sensory in the thickened portions. The thickness of the non-sensory portions of the epithelium is somewhat exaggerated. Sections of decalcified otoliths are shown in Figures 5, 6, 8, and 9.

FIG. 4. Lateral view of right ear to show the positions of sections from which Figures 5 to 10 were drawn.
FIG. 5. Lagena. (See Fig. 4, plane 5.)
FIG. 6. Sacculus and utriculus. (See Fig. 4, plane 6.)
FIG. 7. Common cavity of sacculus and utriculus. (See Fig. 4, plane 7.)
FIG. 8. Sacculus and utriculus, with common cavity. (See Fig. 4, plane 8.)
FIG. 9. Recessus utriculi. (See Fig. 4, plane 9.)
FIG. 10. Anterior ampulla, in a plane parallel with the axis of crista acustica. Axis cylinders are shown penetrating the basement membrane and passing between the basal cells to the zone of distribution between the basal cells and the sense cells.



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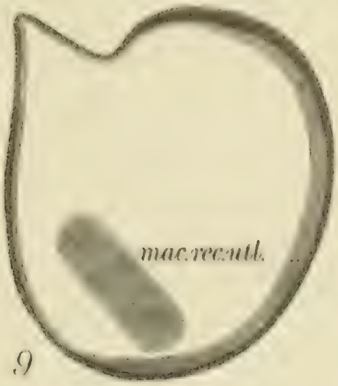

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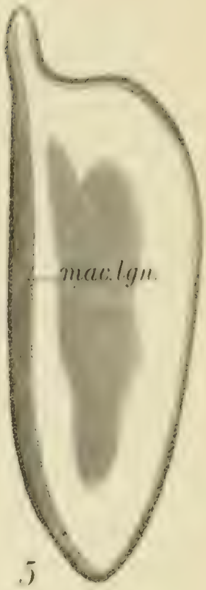

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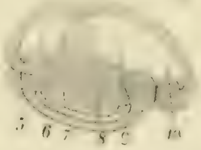

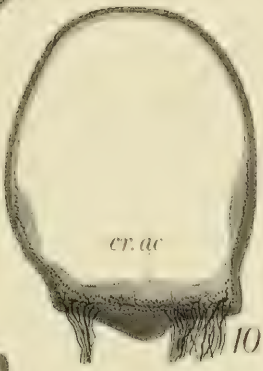

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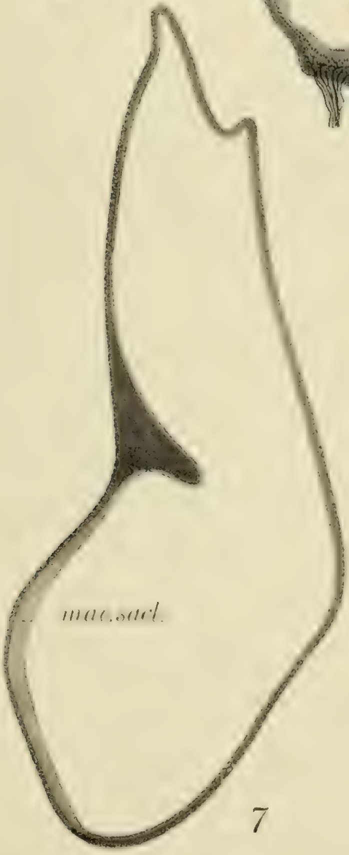

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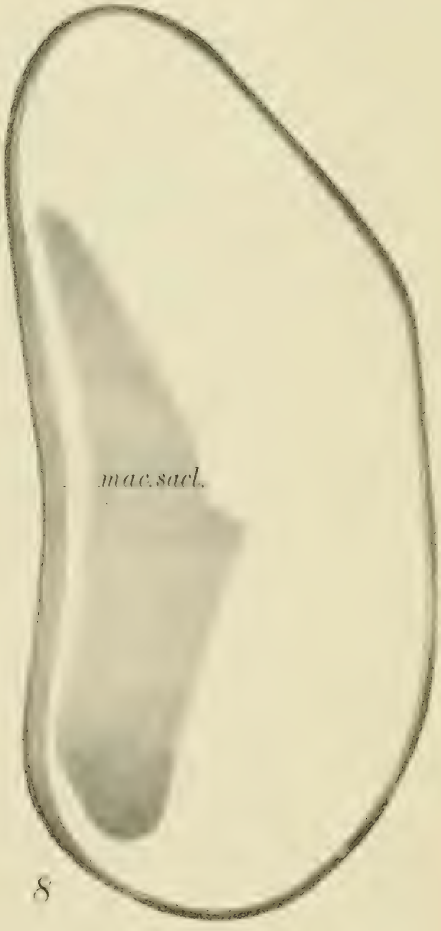

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PLATE 3.

Magnification, 675 diameters.

Portions of cristae acusticae. The sections are at right angles to the free surface of the epithelium which lines the membranous labyrinth. The free ends of the sense cells are above and the basement membrane is below in the drawings of this and succeeding plates.

Figs. 11, 12, 14, and 16. "Kelchbildungen."
Fig. 13. Interlacing axis cylinders in the zone of distribution.
Fig. 15. Intercellular free nerve terminations.

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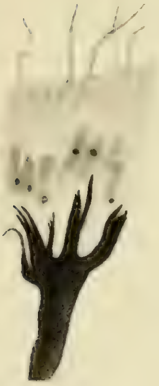

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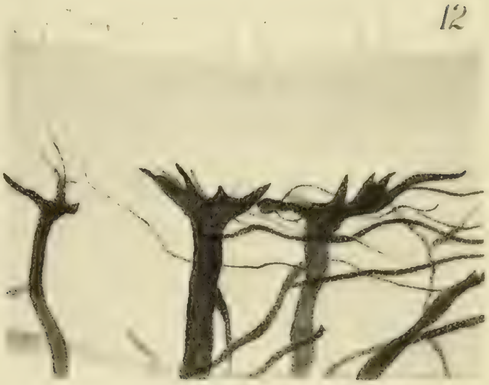

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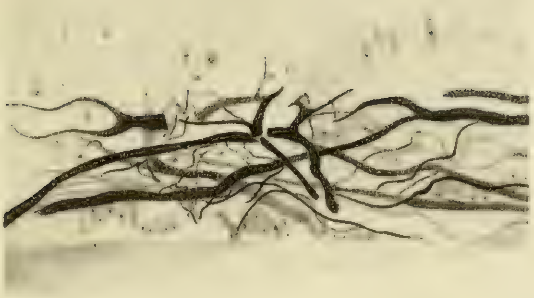

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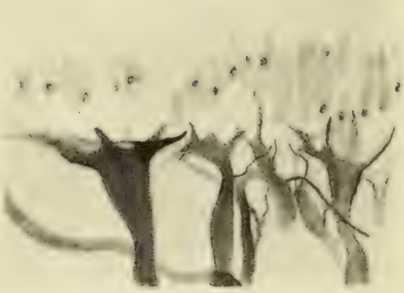

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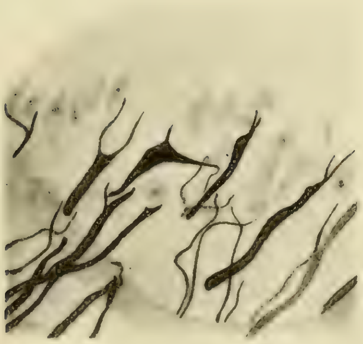

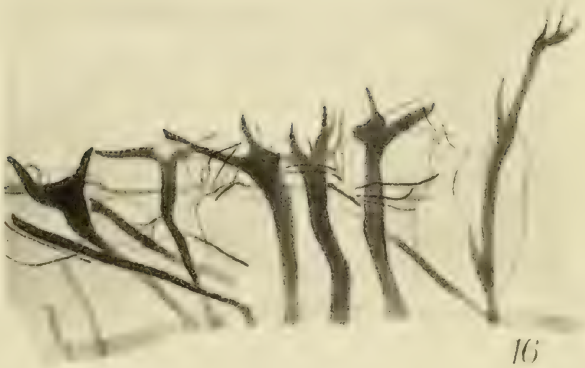

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PLATE 4.

Magnification, 810 diameters.
Details from cristae acusticae.

Figs. 17, 19, 20, 23, 24, 25, 27. Examples of axis cylinders which end free between the sense cells, and near their bases.

Figs. 18 and 26. Instances in which nerve fibres extend nearly or quite to the peripheral ends of the sense cells.

Figs. 21 and 22. Apparent networks of neurofibrillae.

Fig. 26. A free-ending axis cylinder, which extends almost to the cuticula, and is obviously composed of several neurofibrillae.

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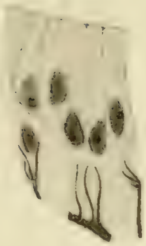

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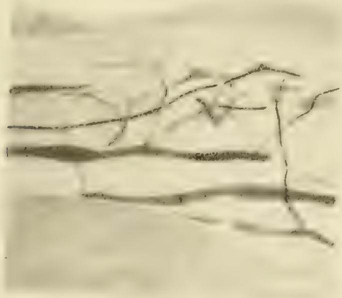

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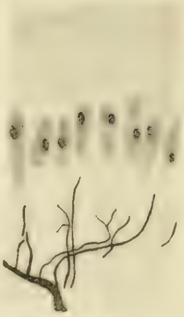

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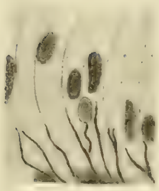

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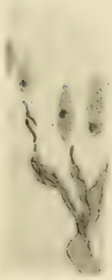

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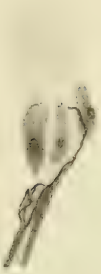

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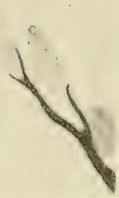

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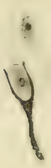

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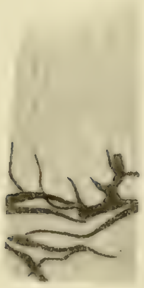

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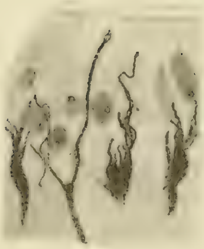

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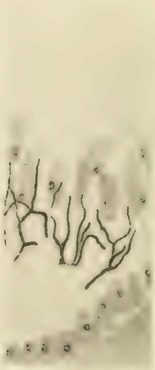

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PLATE 5.

Magnification, 810 diameters.

Sections made in a plane at right angles to the free surface of the epithelium, excepting that from which Figure 39 was drawn.

Figs. 31 and 37. Free nerve terminations in crista acustica.

Figs. 29, 30, 32, and 34. Arborizations of giant fibres in macula acustica sacculi.

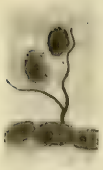
Figs. 35, 36, and 38. Arborizations of giant fibres in macula acustica lagenae.

Figs. 33 and 39. Free nerve terminations in macula acustica sacculi.

Fig. 38. Free terminations in macula acustica lagenae.

Fig. 39. Section in a plane nearly parallel to the free surface of the epithelium in macula acustica sacculi, showing cuticula and intercellular substance in the spaces between the sense cells, which are shown in cross section.

Figs. 38 and 40. Cuticula and intercellular substance in macula acustica lagenae.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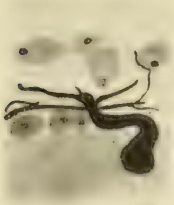
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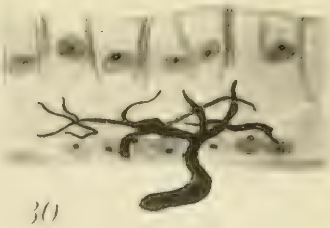
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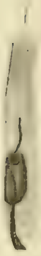

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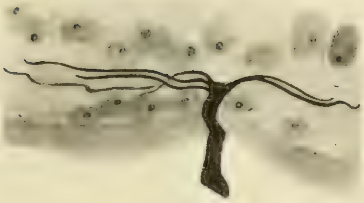

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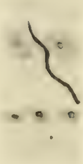

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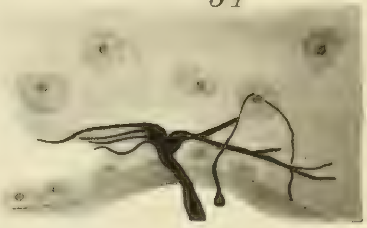

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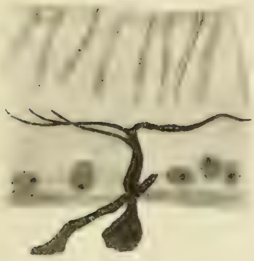

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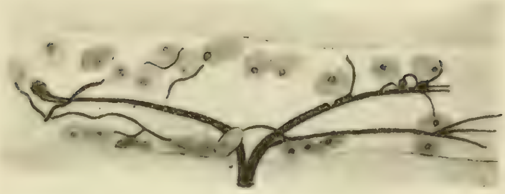

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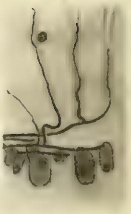

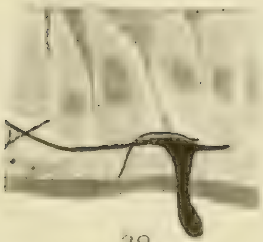
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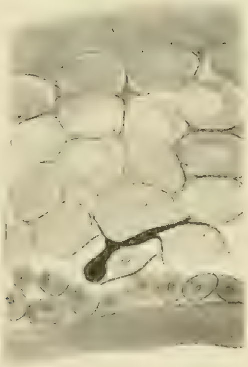
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PLATE 6.

Magnification, 810 diameters.

Figs. 41–43. Nerve terminations in macula acustica neglecta. Sections parallel to the principal axis of the sense cells.

Figs. 44–53. Sections from macula acustica sacculi, in planes nearly parallel to the surface of the epithelium. Sense cells shown in cross section.

Fig. 45. Cytoplasm of sense cells shrunken away from cell walls. Space between sense cells largely occupied by intercellular substance.

Figs. 44 and 46–53. Extracellular axis cylinders.

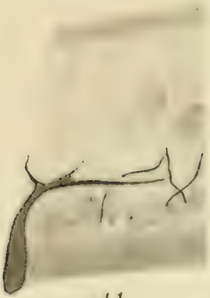
Fig. 44. Cut ends of axis cylinders, between sense cells.

Figs. 46, 47, 50, 51. Small axis cylinders in close proximity to surface of sense cells.

Figs. 48, 50, 51. Axis cylinders bifurcating at surface of sense cell and sending a branch to either side of it.

Fig. 52. Tangled intercellular axis cylinders, which presented, when examined with low power dry lens, the appearance of being anastomosed to form a closed network; but when studied under high-power oil-immersion lens were found to be distinct.

Fig. 53. Two of the sense cells contain filaments which present the appearance of nervous matter.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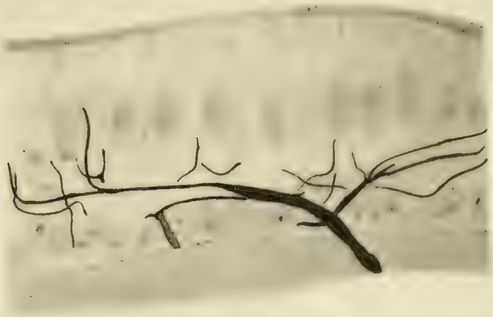
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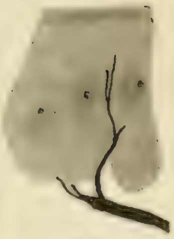
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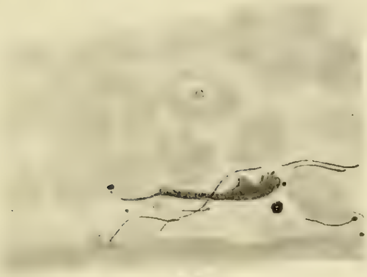
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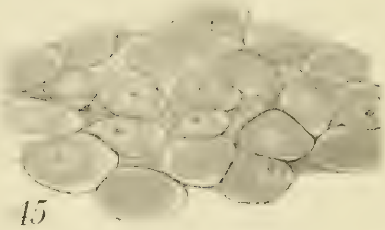
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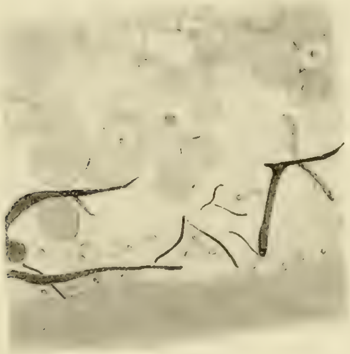
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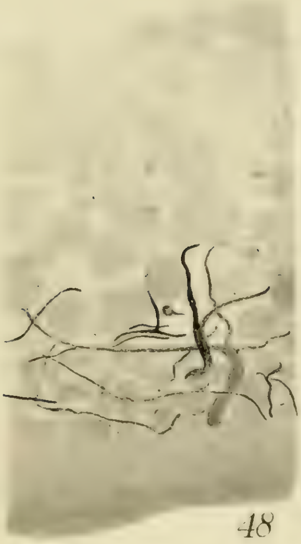
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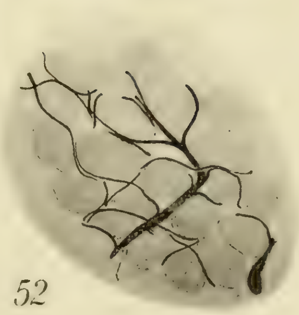
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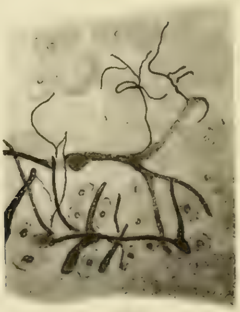
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Bulletin of the Museum of Comparative Zoölogy
AT HARVARD COLLEGE.
VOL. LIII. No. 5.

THE REACTIONS OF AMPHIBIANS TO MONOCHROMATIC LIGHTS OF EQUAL INTENSITY.

BY HENRY LAURENS.

CAMBRIDGE, MASS., U. S. A.:
PRINTED FOR THE MUSEUM.
JUNE, 1911.

No. 5.— CONTRIBUTIONS FROM THE ZOÖLOGICAL LABORATORY OF THE MUSEUM OF COMPARATIVE ZOÖLOGY AT HARVARD COLLEGE. E. L. MARK, DIRECTOR. No. 223.

The Reactions of Amphibians to Monochromatic Lights of Equal Intensity.

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I. Introduction.

Among the numerous papers concerning the reactions of animals and plants to light that have appeared in recent years, there are a considerable number of references to the reactions of organisms to different colored lights, though there are comparatively few papers which deal with these reactions alone. What work has been done on testing the reactions of organisms to different colored lights, however, cannot be considered as very fundamental. This is due to the fact that, in the majority of cases, the colored lights were obtained by filtering white light through colored media, either glass or colored solutions. This method of obtaining colored light has recently been shown to be practically worthless, for even the best of such screens also let through a large amount of dark heat rays. It is therefore impossible to state whether the reactions that occur under such conditions are to be attributed to the visible or to the invisible energy.

Some little work has also been done with spectral light, but the results obtained by this method are hardly to be considered more valuable than those obtained by the use of screens, for the reason that no effort was made to render the lights used equal in the amount of radiant energy that they contained. If the curve representing the relative amount of radiant energy contained in the different regions of the prismatic spectrum of a common source of light be plotted, it will be seen to begin very low in the blue, and rise gradually until it reaches its maximum in the red. Some investigators have attempted to make what they considered the intensities of the different regions equal by making the brightness of the different lights, as judged by the human eye, the same. But this procedure is inaccurate, for the effect of the different lights on the human eye is not proportional to their energy content. Even the effect on the human eye varies with the amount of illumination, for in a brilliant spectrum the maximum brightness of the spectral colors is in the yellow, while in feeble illumination this shifts to the green. The changing luminosity and color value of light with changes of light intensity — the Purkinje phenomenon — is a clear indication that the relative brightness of the spectral colors to lower organisms is probably very different from the condition found in the human eye, and that the human eye, therefore, cannot be employed as a means of measuring the relative intensities of light to be used in biological work.

There is only one piece of investigation, so far as I know, in which any attempt was made to measure the relative intensities — and by this I mean the energy contained in each light — of different colored lights by any physical apparatus. Kniep and Minder (**:09**), working on the assimilation curve of chlorophyl-containing plants, came to the conclusion that the reason there were so many conflicting opinions on this subject was that not enough attention had been paid to the relative intensities of the different colored lights. They therefore proceeded to measure the absolute energy of sunlight in the different spectral regions by means of a Ruben's thermopile, in connection with which was a very delicate galvanometer, and to adjust the light until the radiant energy in the different colors was the same for each region. The value of this work is unfortunately much lowered by the fact that the experimenters used screens to obtain their different colored lights, and therefore the dark heat rays must have been present, and have had their effects.

I shall attempt to summarize, as briefly as possible, the results of the work that has been done on testing the reactions of amphibians to colored lights.

The first investigator, to my knowledge, to test the reactions of amphibians to different colored lights was Graber, who in two publications (**'83** and **'84**) made an exhaustive series of experiments in an attempt to solve the problem as to whether, and to what extent, animals were able to distinguish intensity and color differences. In all of his work, Graber used colored glasses to obtain his different lights, two colors being used in such a way that the animal was given the choice of two compartments, each illuminated by a different colored light. He tested three species of amphibians, the first of these being the salamander, Triton cristatus. This he found (**'83**, p. 220, and **'84**, p. 108) to be strongly negatively phototropic in white light, both in the normal and eyeless condition, though less so in the latter. When Triton was given the "choice" between two different colored lights, Graber found that it "preferred" the less refrangible rays, i. e., the red. This was so for both normal and eyeless individuals. He was sure that these reactions were not due to the influence of temperature, since he used a heat screen for the blue, and none for the red light, and obtained similar results, which, since the salamander is "thermophob," showed that heat was not a determining factor in the reactions. He also (**'84**, p. 120) found that Rana esculenta, which, according to him, was negatively phototropic in white light, when exposed to different colored lights, showed a "preference" for red, as compared with blue and green, and that green was "preferred" to blue. He did not experiment with blinded frogs. With the toad, Bufo vulgaris, he (**'84**, p. 124) found that, while it, too, was negative in white light, it "preferred" blue to red, and green to red.

Loeb (**'90**, p. 89) intimated that the frog was negatively phototropic in white light. As to colored lights, he obtained the same results, no matter through what colored medium the light was passed, there being a difference only in the intensity of the stimulation, that is, the difference was only quantitative. The stimulating effect of the more refrangible rays, he found to be much stronger than that of the less refrangible. This he showed by exposing a frog at the same time to more and less refrangible rays whose directions were opposite. Under these circumstances, only the strongly refrangible rays exerted a directive influence on the animal, one end of the receptacle in which the frog was placed being of blue glass, and the other of red. The animals then behaved as if only the light from the blue glass were falling on them, and moved quickly away from it. Loeb did not mention whether he used any intermediate colors.

Dubois (**'90**, p. 358) found that the blind Proteus, which has rudimentary eyes, was strongly negatively phototropic. On exploring the whole surface with a small beam of light, he found that the sensitiveness to light was distributed over all of the body, but that the tail and the head were the most sensitive regions. He then covered the eyes with gelatine and lamp-black, and again obtained negative responses to the light. He placed a heat screen between the light and the animal, so that he was certain that heat had nothing to do with the reactions. Dubois thus showed that while Proteus was sensitive to light through the skin, it was not as much so as through the eyes. Colored glasses, transmitting lights that were non-monochromatic, were used in testing the reactions to colored lights. Dubois found that Proteus showed least sensitiveness to yellow light, and that the sensitiveness to the other lights was in the following decreasing series: violet, blue, red, green. When the eyes were covered, the results were very inconstant. If Proteus was experimented with according to Graber's method of placing animals in a chamber with an opportunity to choose between two lights, as was done for the blinded Triton, Dubois found that in the absence of darkness it went most quickly into yellow and red light. The lights according to the rapidity with which Proteus would go into them were as follows: red, yellow, green, violet, blue, white. The intensities of the lights ran in the following series: yellow, blue, red, green, violet.

Torelle (**:03**) showed that the frogs Rana virescens virescens and R.

clamata, which were positive in white light, when exposed to different colored lights obtained by passing white light through colored solutions, turned away from red light, and moved toward blue light; that they also moved toward green and yellow light, but were not oriented by either. When given the choice between red and green light, which were admitted at opposite ends of a receptacle, the frogs moved away from the red to, or toward, the green. When given the choice between red and yellow, they moved away from red to yellow; and when given the choice between red and blue, they moved immediately toward the blue.

Reese (:06) found both Necturus and Cryptobranchus to be negative to white light, the head of Necturus and the tail of Cryptobranchus being the most sensitive regions. In both of these forms, blue light was found to be more effective in bringing about responses than was red.

Holmes (:07, p. 349), in discussing the light reactions of frogs, said, "in all animals thus far investigated it is the blue and violet rays that are the most influential in evoking the phototactic response; the effectiveness of the other colors of the spectrum diminishes in order from blue to red. If frogs are placed in a box illuminated through one end with blue light, and through the other with red, they soon gather at the blue end. If they have the choice between yellow and green, they go toward the green; in general it may be said that where they are able to go toward one of two colors of equal intensity they move to the color lying nearest the violet end of the spectrum."

Eycleshymer (:08, p. 304), while rearing larvae of Necturus, placed strips of black, white, red, yellow, green, and blue paper beneath the glass aquaria in which they were kept. At first, he could observe no "preference" for one color over another, though later he obtained evidence "that by far the highest percentage of larvae were found over the green, whether this was placed on the side of greatest or least diffuse daylight." Decapitated larvae "were most frequently found on the colors in the half of the spectrum toward the violet end."

Pearse (:10, p. 176) found that the toads Bufo fowleri, and B. americanus were positive in white light, in the eyeless as well as in the normal condition; but, when they were tested in colored lights obtained by passing white light through colored solutions (p. 187), the reactions did not agree with those of normal frogs. When Rana palustris, in the normal condition (p. 189), was tested, blue light was found to be most effective in the production of positive responses, and red the least, with a gradual decrease in effectiveness between them. But when eyeless toads were tested (p. 191), they were equally positively phototropic in all the lights used. To quote (p. 206), "it may be said that, while both the skin and eyes are sensitive to the whole range of the visible spectrum, color-sensitiveness is present only in the latter."

The results thus summarized seem to point, therefore, to the view that the blue end of the spectrum is the most effective in the production of phototropic responses, and that this is true for both positively and negatively phototropic forms. The results obtained by Graber on the toad, and by Dubois on Proteus, do not agree with this. But there is no certainty that any of the results obtained by the earlier investigators in this subject are due unquestionably to a response to difference in color, or wave lengths, as such, rather than to the difference in intensity, or to the radiant energy content, of the several lights.

The experiments described in the present paper were carried out (1) to ascertain whether amphibians showed a sensitiveness to lights of different wave lengths, exclusive of their intensity value; and (2) to decide whether this sensitiveness, if present, resided in the eye, in the skin, or in both the eye and the skin.

It is a pleasure to acknowledge my gratitude to Prof. G. H. Parker, under whose direction the work was undertaken, and without whose untiring interest, suggestions, and criticism it would not have been accomplished. I take this opportunity also to express my thanks to Mr. A. O. Gross, a student in the Laboratory, for invaluable assistance in the construction of some of the details of the apparatus.

II. Methods.

1. Apparatus.

The apparatus used in these experiments is based on appliances worked out by G. H. Parker and E. C. Day, an account of which is shortly to be published. The plan of the apparatus is shown in Figure 1. It consisted of two light-generators, *A* and *B*, so placed, with reference to each other, that the light from *A* entered a dark chamber (*L*), opposite that from *B*; and of a table (*T*), covered with a slab of slate, on which the animals whose reactions were to be tested were placed.

A. The Light-Generator.

The light-generator may be described as follows: the sources of light were Nernst glowers on a 220-volt circuit, the light from which passed, first through a rectangular opening in a diaphragm of blackened sheet iron (*C*), then through a converging lens (*D*), which was at such a distance from the source of light that the conjugate foci were at equal distances from the lens, and finally through a large glass prism (*F*) filled with carbon bisulphide, placed within the focal distance of the lens, and at the angle of minimum deviation. The spectrum thus obtained was cut down by diaphragms of blackened cardboard with narrow vertical slits of appropriate size and position. These diaphragms were placed in a holder (*G*) at the focal points of the several lights used. Side reflections were eliminated by enclosing light, lens, prism, etc., in a covered box (*H, I, J, K*), which was blackened inside, and completely closed except at the end farther from the source of light, where the light from the prism was projected into the dark chamber. Every care was taken, by the use of suitable screens, etc., to exclude from the dark chamber all light except that proceeding from the prism. It was possible to revolve the generator on a pivot at *P*, so that the direction of the light could be changed within certain limits. An adjustment at *X* enabled the experimenter to change slightly sideways the position of the lamp. The colored lights used in the experiments were four in number, as follows: blue, 420–480 $\mu\mu$; green, 490–550 $\mu\mu$; yellow, 570–620 $\mu\mu$; and red, 630–655 $\mu\mu$. The terms blue, green, yellow, and red are used for convenience in designating these lights, and not in their strict physical application.

B. The Combined Apparatus.

The complete apparatus, as shown in Figure 1, consisted of two such light-generators, placed at opposite sides of the dark chamber (*L*), into which the light from each was projected, passing first through a plate-glass window (*M*) in a screen (*N*), the window (*M*) being of similar size and thickness to that in the box which surrounded the radiomicrometer by which the light was originally measured. The walls and ceiling of the dark chamber were of opaque black cloth, there being an aperture (*O*) through which the observations were made. The height of this dark chamber was 70 cm., and its floor area 130 ×

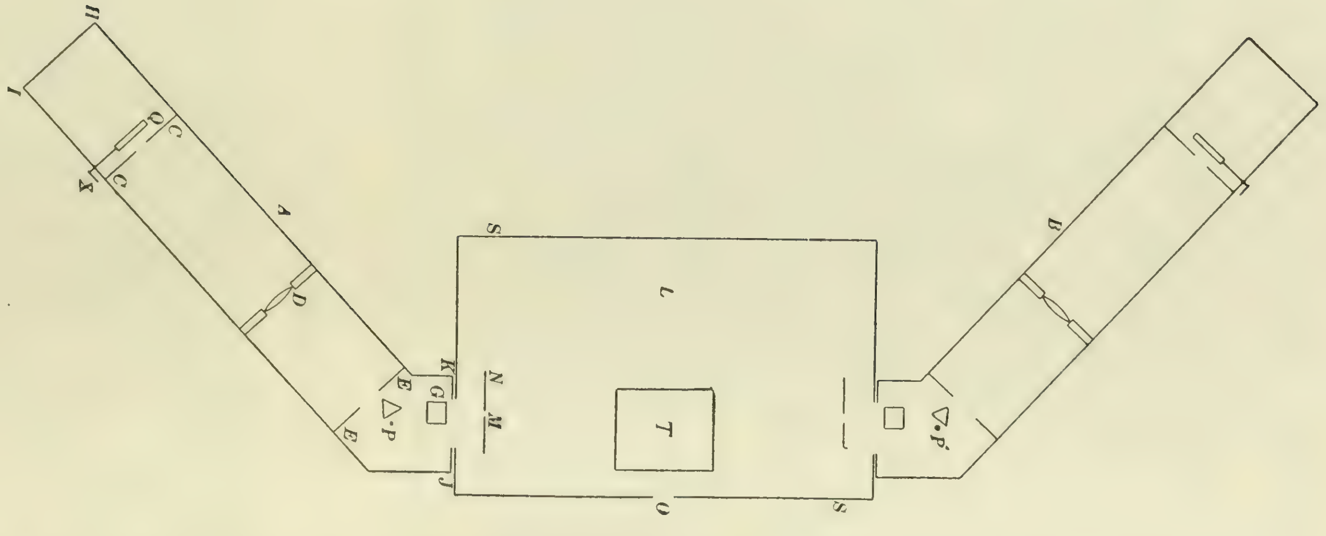

FIG. 1. Plan of apparatus for testing the reactions of toads to monochromatic lights. *A*, generator *A*; *B*, generator *B*; *CC*, diaphragm of blackened sheet iron; *D*, converging lens; *EE*, diaphragm; *F*, prism; *G*, diaphragm-holder; *HIJK*, covered box enclosing light, lens, prism, etc.; *L*, dark-chamber; *M*, plate-glass window; *N*, screen; *O*, aperture in screen through which the observations of the reactions were made; *P* and P^1, positions of pivots on which the boxes could be revolved; *Q*, lamp; *SS*, screens of black cloth; *T*, table on which the toads were placed; *X*, adjustment for changing sideways the position of the lamp.

80 cm., thus giving ample space for work. The experiments that were made to test the reactions to lights of different wave-lengths may be divided into two main sets; (1) Reactions to single monochromatic lights, and (2) Reactions to balanced pairs of monochromatic lights. In the first, the animal to be tested was exposed to a single light, which impinged upon its side at right angles to its long axis. In the second, the animal was placed midway between two lights of different, or of the same, wave-lengths, which impinged at right angles upon opposite sides of its body. A rather full description of the way in which the combined apparatus was used in making tests of the reactions will not be out of place.

In preparing the apparatus for tests with single monochromatic lights, the following steps were taken: the four lights used (p. 259) were balanced, that is, made equal in the energy they contained, by adjusting the diaphragm openings (p. 259) one after another, till they gave equal deflections in a radiomicrometer. This was done only for the lights in generator *A*; for convenience these lights will be called the standard lights. From the measurements with the radiomicrometer, it was found that, in order to procure four colored lights that would be equal in the energy they contained, it would be convenient to use three forms of lamps, these lamps differing, however, only in the number of glowers employed in each. The lamp used for obtaining red light contained a single glower; that for yellow, two glowers; and that for the green and the blue, three glowers. A separate cardboard diaphragm, in the holder *G*, with a vertical slit of a different width and position for each light was also necessary. These diaphragms were set in position accurately by means of a spectroscope, the necessary range for each light having been worked out experimentally. The lights from generator *B* were now adjusted so as to cover the same spectroscopic range as those from generator *A*. In this way the two lights were not only made equal spectroscopically, but they were also equal in intensity. This last fact was clearly demonstrated when a Lummer-Brodhun photometer was placed midway between two lights of the same wave-lengths proceeding from the two generators, the difference in intensity of the two lights being exceedingly small.

Perhaps all this can be made clearer by a concrete case. Suppose, for example, that it was desired to test the reactions in blue light. The procedure was as follows: The 3-glower lamps were placed in both boxes and lighted, the appropriate diaphragms were next placed in position, and the wave-lengths of the light proceeding from generator *A* read on the spectroscope, the adjustment at *X* (Fig. 1) being used to shift the lamp until the wave-lengths read 420–480 $\mu\mu$. This was then repeated for the light proceeding from generator *B*. From what has been said, the radiant energy in the blue light from generator *B* must now be approximately equal to that in the blue light from generator *A*. The boxes were then so revolved on the pivots (*P* and *P′*, Fig. 1), that the light rays from the two were parallel and opposite. A photometer placed midway between the two boxes in the path of the lights then showed that the intensity of the two lights was equal. The lights were now in such a condition that the animals could be introduced for tests. Under the separate sections the special methods used and the procedure of the tests themselves will be more fully given.

It may seem to have been unnecessary to have gone to all of this trouble, when the wave-lengths of the lights from generator *A* could have been read, and the animals tested in this. But the use of two lights of the same wave-lengths from opposite sources, lessened the chance of experimental error. Under the description of the reactions to single monochromatic lights, it will be shown how the method of rotation and the use of the two lights from opposite sources were combined to offset the influence of compensating movements upon the reactions. The procedure for the blue, just described, was repeated for each of the other three sets of colored lights.

When the reactions to balanced pairs of monochromatic lights were tested, the procedure, while in the main the same as that just described, differed somewhat from it. Here there were two lights from different sources, midway between which the animal was placed, so that the lights impinged at right angles upon opposite sides of its body. Further, the lights were of different wave-lengths, except in a few experiments that were carried out to check the equality of the lights from the two light-generators. For the sake of clearness in explaining the procedure, a concrete case again will be taken. Suppose, for example, that it was desired to test the reactions of a lot of toads in blue and in red light. The single-glower lamps were placed in both boxes, lighted, and the appropriate diaphragms placed in position. The light proceeding from generator *A* was then scrutinized in a spectroscope, and the lamp adjusted until it read 630–655 $\mu\mu$, which were the wave-lengths of the red light used. The same process was repeated for generator *B*. The energy contained in the two red lights was then assumed to be the same. The boxes were then so revolved on the pivots (*P* and *P′*, Fig. 1), that the light rays from the two were parallel and exactly opposite. A photometric reading showed that the intensity of the two lights was approximately equal. The lights in generator *A*, it will be remembered, were standard lights of equal intensity, these being the ones measured with the radiomicrometer, and all that has been just described was done to make sure that the red light in generator *B* was similar in energy content, intensity, to that of the standard in generator *A*. The 3-glower lamp was then substituted for the 1-glower lamp in generator *A*, and the light adjusted until it was found to be 420–480 $\mu\mu$, as read in a spectroscope. The boxes were then again so revolved that the lights were exactly opposite. It is clear that under such circumstances the radiant energy contained in the red light in generator *B* must be equal to that contained in the blue light in generator *A*. The lights were now in such a condition that tests of the reactions of the animals could be made, the two lights being of different wave-lengths, but containing the same amount of radiant energy. The special methods used, and the procedure followed, will be more fully considered under the section devoted to the reactions to balanced pairs of monochromatic lights.

2. Material.

The common toads, Bufo americanus and B. fowleri, were the forms employed in all of the experiments described in the succeeding pages. No separate records were made of the reactions of the two species, and there was no reason for believing that they were in any way different. Most of the toads used were collected in the vicinity of Cambridge, Massachusetts, but a few were procured from Orlando, Florida. The stock was kept in a basement room, in boxes, the floors of which were covered with about eight inches of earth. When a lot of toads was selected for experimentation, each individual was kept by itself by being placed alone in a medium-sized battery-jar, the bottom of which was covered with earth. In this way, it was possible to keep track of the individual's reactions during a long series of experiments. While the experiments were in progress, the toads were periodically fed meal worms, which they ate readily.

The experiments were carried on in the spring months of 1910, and the fall and winter months of 1910–1911. They were all performed in an experimental dark-room in the basement of the Museum. The temperature of the room varied between 19° C. and 24° C., though it was only occasionally that it went as high, or as low as the extremes given, the average temperature being between 20° C. and 21° C.

III. Observations.

1. Reactions to Single Monochromatic Lights.

A. Reactions with both the Skin and the Eye as Receptors.

In testing the reactions of toads to single monochromatic lights the combined generators, as described on p. 261–262 were used. These generators were so adjusted as to give out lights of the same wave-lengths, and screens were so arranged that one or the other light could be cut off in the dark chamber. The lights employed were those already described on p. 259.

Twelve toads were successively tested in each light, and each animal was given eight trials, of which four were made in the light from one generator, and four in that from the other. The total number of trials in each light was therefore 96. The twelve toads were always brought into the experimental dark-room the afternoon before the experiments were made, and care was taken that they should not in any way be illuminated until they were exposed to the light in which they were to be tested. They were thus subjected to a condition of dark for at least fourteen hours before being tested, and at the time of the test they were undoubtedly "dark adapted." After a toad had been tested in a certain light, it was again placed in the dark, and was not experimented with for at least an hour, after which period its reactions in another light were tested. I do not think that there can be any doubt that the toads were thus always "dark adapted" for each set of tests.

The procedure used in the experiments was as follows: the lights from both generators were tested spectroscopically and photometrically; after they had been found to be equal, that from generator *B* was screened off, and that from generator *A* admitted to the dark chamber. Toad No. 1 was then placed on the table, in the beam of light from generator *A*, and held with its head toward the observer. Before any reaction could be made, it was quickly rotated through 180 degrees, so that its head was directed away from the observer, and it was freed in this position, with the light impinging on its left side and at right angles to its long axis. At each of the succeeding trials, the toad was rotated clockwise, so that for the second trial it was headed toward the observer, with the light impinging on its right side; for the third, away from him, etc. After a total of four trials had thus been made, the light from generator *A* was screened off, and that from generator *B* was admitted to the dark chamber. The toad was then subjected to a second set of four trials, which were carried out in a similar way to the first set, except that the toad was rotated counterclockwise. Toad No. 2 was next taken in hand, and the same procedure was carried out with it as with toad No. 1, but with this difference, that with No. 2 the light for the first four trials came from generator *B*, and for the last four from generator *A*, instead of the reverse. The procedure with toad No. 3 was similar to that with toad No. 1, and with toad No. 4 to that with No. 2, and so on through the series of twelve toads. In this way the right and left sides of the toads were alternately exposed to the lights, and to the lights from both generators. It is well known that frogs and toads will respond to a sudden turning in one direction by a compensating movement of the head in the opposite direction; the method of rotation above described offsets any influence of these compensating movements upon the reactions. It may be here mentioned that no toad was ever under actual experimentation for more than forty minutes on any day, and very seldom for more than five minutes continuously.

A period of five minutes was given each toad in which to react. If, after this period had elapsed, the toad had shown no response, it was rotated through 180 degrees to head in the opposite direction for the next trial. The reactions were, however, usually very quick and definite. I obtained reactions, occasionally, in less than ten seconds after a toad had been placed in position for a trial, though the average time of the reactions was a little less than a minute. The nature of the reactions was as follows: The toads turned until they headed toward or away from the light, and then hopped in the given direction. With some toads the whole response of turning and hopping was very quickly accomplished, a sudden turn being followed by as sudden a hop. In others, there was a sudden turn followed by a hop only after an appreciable length of time had elapsed. In still others, a considerable period passed before even the turning toward or away from the light took place. In a few instances, a slight mechanical stimulation, such as a delicate touch from behind, was required to elicit the first response, and then the animals eventually crawled, rather than hopped. There were a very few cases in which toads turned toward or away from the light, but made no further movements. All such forms of movement were recorded as either positive or negative reactions, but there was still another form of reaction, which I have called "indifferent," that is, locomotion straight ahead, without apparent reference to the light.

There was some slight inconstancy in the reactions of the same and of different individuals. A given individual seldom showed consistently the same percentages of responses to the different lights in the different sets of trials. This inconstancy in the reactions was very noticeable in not more than two or three individuals, though it was very slightly present in several. The precise factors upon which this inconstancy depended were not ascertained, but they were probably functions of the physiological states of the animals, changing slightly from time to time, and therefore changing the nature of the response in any given individual.

The results of the first series of tests are given in Table 1. Three sets of twelve toads each were tested in the way described, the total number of trials for each light being, therefore, 288. The same twelve toads were used for the first two sets, and the tests for these followed immediately upon each other. A second lot of twelve toads was, however, selected for the third set of tests. The results with this second lot of twelve toads were so similar to those obtained with the first, that they are not given separately in the Table, but together with the other two sets.

It will be seen, by referring to Table 1, that all the lights used produced more positive responses than negative. Thus, in the blue light, out of 288 trials, 251 were positive; in the green, 230; in the yellow, 194; and in the red, 167. A comparison of the effectiveness of the four lights can be made more easily if the percentages of positive responses are considered, rather than the actual numbers. This effectiveness was greatest for the blue light (87 %), and decreased for the other lights in the order of the spectrum. The green light (80 %) was, however, not very much less effective than the blue. The decrease in effectiveness between the green and yellow lights was greater than that between the blue and green, the number dropping in yellow to 67 %. The red light was again less effective than the yellow, the percentage of positive responses being only 58 for this light. This percentage of positive responses in red light, if decreased only 8, would make the percentage of positive and negative responses, under this stimulus, equal; that is, the percentage of movements toward the dark would be as great as that toward the light. The fact that there were 58 % of positive responses, and only 42 % of negative responses, shows clearly, however, that the red light must be regarded as more effective than darkness in the production of responses in toads.

TABLE 1.

Reactions of toads to monochromatic light received through both eye and skin.

Monochromatic lights of equal intensity		Blue 420–480 μμ				Green 490–550 μμ				Yellow 570–620 μμ				Red 630–655 μμ			
Reactions	Directions of	+	—	±	O	+	—	±	O	+	—	±	O	+	—	±	O
	Number of responses	251	37			230	58			194	94			167	121		
	Percent of responses	87	13			80	20			67	33			58	42		

The numbers under + indicate total numbers of reactions toward the light; under—, away from the light; under ±, without reference to the light (indifferent); under O, no reaction within five minutes.

By way of summarizing the results of the experiments with single monochromatic lights in which both the eye and skin acted as receptors, it may be stated that all four colored lights used produced positive responses. Blue light was the most effective, and the other lights formed a decreasing series, corresponding roughly to their relative position in the spectrum, the red light being but slightly more effective than darkness.

B. *Reactions with the Eye as Receptor.*

It was natural to suppose that the reactions of toads with both the eye and skin acting as receptors, were dependent upon the eye, but it was conceivable that they might also depend in some measure upon the skin. The skin of many amphibians has been found to be very sensitive to white light. Parker (:03) showed this to be the case in the frog, Rana pipiens. He also reviewed the literature of the subject of the sensitiveness of the skin to stimulation by light, pointing out that similar results had been obtained by previous observers on Triton and Proteus, among amphibians, as well as on certain fish and other metazoans. Later (:05) he showed that the skin of Ammocoetes also possessed this sensitiveness to light, the tail being the most sensitive region. Reese (:06) obtained similar results with Cryptobranchus, but he found that in Necturus when illuminated from above the head was the most sensitive part, due to stimulation received through the eyes; but that, when the ventral surface was illuminated, the head was less sensitive than the tail. Payne (:07) showed that sensitiveness to light was present in the skin of the blind fish Amblyopsis, and that "they seem to be equally sensitive on all parts of the body." Eycleshymer (:08) found that the decapitated larvae of Necturus oriented to light in the same manner as the normal larvae do, though they turned as frequently from the light as toward it, while normal larvae turned usually toward the light. After decapitation, the tail was the most sensitive region. More recently Pearse (:10) has found that, out of nine species of amphibians, comprising anurans and urodeles, which he tested, seven "after the removal of their eyes gave photic responses which were like those of normal individuals." The toad was one of the seven species in which Pearse found the skin to act thus as a photoreceptor. It was therefore natural to expect that the skin of the toad would show a sensitiveness to lights of different wave-lengths. Hence it seemed desirable to test toads in single monochromatic lights; first, with only the eye as receptor; and secondly, with only the skin as receptor. In testing the reactions with only the eye as the receptor, the method and procedure were the same as described on p. 264–266, except that the beam of light was made to pass through a small opening in a screen placed close to the table (T), fig. 1, the opening being made of such size, and so adjusted, that only the eye of the toad and a very small area of skin around it were illuminated.

The results of these tests are given in Table 2. Again three sets of twelve toads each were tested, the total number of trials in each light being, therefore, 288. For all three sets, the same twelve toads were used that had been used in the first two sets of experiments, in which both the eye and the skin served as receptors. Of the original twelve, however, two individuals died in the course of the experiments, and their places had to be supplied by two other toads. The nature of the reactions was the same as when both the eye and the skin were exposed. The toads, after they had turned toward the light, moved toward it, so that the beam of light was kept on the eye.

It will be seen, by referring to Table 2, that the reactions of the toads when only the eye was exposed to the light were essentially the same as when the whole body was exposed, and that the sequence of the lights, as determined by their effectiveness in stimulating the toads,

TABLE 2.

Reactions of toads to monochromatic light received through the eye only.

Monochromatic lights of equal intensity		Blue 420–480 μμ				Green 490–550 μμ				Yellow 570–620 μμ				Red 630–655 μμ			
Reactions	Directions of	+	—	±	O	+	—	±	O	+	—	±	O	+	—	±	O
	Number of responses	246	42			214	74			187	101			160	126		2
	Percent of responses	85	15			74	26			65	35			55	44		1

The numbers under + indicate total numbers of reactions toward the light; under—, away from the light; under ±, without reference to the light (indifferent); under O, no reaction within five minutes.

corresponded to the sequence in the spectrum with blue as the most effective stimulus, and red as the least. There were only 55 % of positive responses in red light, 44 % of negative, and 1 % of no reaction, within five minutes. A decrease of 5 % would therefore make the percentage of positive responses equal, under this stimulus, to the sum of the percentages of negative responses and no reactions. While, therefore, the red light, owing to the presence of the 55 % of positive responses, must be regarded as more effective than darkness in the production of responses, it cannot be regarded as much more so.

By way of summarizing the results of the experiments with only the eye as receptor, it may be said that they were essentially the same as when both the eye and skin acted as receptors. Blue light was the most effective stimulus in the production of positive responses, and the other lights formed a decreasing series, following the sequence in the spectrum, red light being only slightly more effective than darkness.

C. *Reactions with the Skin as Receptor.*

Having demonstrated that the eye was concerned in the reactions in which the whole body was exposed to the light, it remained to test the skin in this respect. To do this, it was necessary that the eye be protected from illumination. Hoods, made from the finger-tips of fairly heavy rubber gloves, were placed over the snouts of the toads in such a way that the eyes were completely covered. These hoods were attached by two threads to a light copper wire, which encircled the animals' bodies just back of the fore-legs. To insure that no light penetrated these hoods, they were blackened inside and out with India ink. The extreme tip of the hood was cut off so that respiration should not be interfered with.

The hoods were at first a source of some irritation to the toads, which struggled more or less to get rid of them. In a day or two, however, the toads showed no inconvenience when the hood was slipped on, but remained quietly seated on the bottom of the battery-jars in which they were kept (p. 263). In testing the reactions, the methods and procedure of the experiments were the same as described on p. 264–266.

The results of these tests are given in Table 3. Four sets of twelve toads each were tested, the total number of trials for each light being, therefore, 384. For the first two sets, six of the twelve toads were the same as had been used in the first two sets of experiments in which the eye and skin acted as receptors, and in the three sets of experiments in which the eye acted as the receptor; the other six were toads which were supplied to take the place of six that had died during the course of the experiments. For the last two sets, two new lots of twelve

TABLE 3.

Reactions of toads to monochromatic light received through the skin only.

Monochromatic lights of equal intensity		Blue				Green				Yellow				Red			
		420–480 μμ				490–550 μμ				570–620 μμ				630–655 μμ			
Reactions	Directions of	+	—	±	O	+	—	±	O	+	—	±	O	+	—	±	O
	Number of responses	296	88			271	113			248	136			217	166	1	
	Percent of responses	77	23			71	29			65	35			57	43		

The numbers under + indicate total numbers of reactions toward the light; under—, away from the light; under ±, without reference to the light (indifferent); under O, no reaction within five minutes.

toads each were selected. These tests were carried out, therefore, on thirty-six separate toads. It should be mentioned that the first series of reactions of hooded toads which were recorded were so irregular that I was led to believe that the skin of the toad was not sensitive to differences in wave-lengths. This was absolutely disproven by the later series, as will be seen by referring to Table 3. The irregularity in the reactions of the first set were probably due to the irritation caused by the hoods. This first series of trials, which was carried out relatively soon after the hoods had been placed on the toads, but after they had apparently become used to them, were therefore thrown out, and in all the later experiments a toad was not tested until it had undergone the experience of wearing a hood for a few hours every day for a week. Moreover, in the immediate preparation for the experiments, the twelve toads were hooded the afternoon before the tests, and were left so in the experimental dark-room over night. On two or three occasions, a toad was found in the morning with the hood off. In such cases the hood was replaced, exposing the toad as little as possible to light, and this animal was not tested until it had been for at least 45 minutes in the dark.

It will be seen, by referring to Table 3, that the reactions of toads, when only the skin was exposed, were in general the same as when the whole body, or only the eye, was exposed. All the lights produced positive responses, but the effectiveness of the blue was the greatest, and that of the other lights decreased in the order of the spectrum. The green light, however, was not very much less effective than the blue, there being a difference in the number of reactions of only 6 % between them; nor was the yellow much less effective than the green, the difference being again only 6 %. The red was clearly less effective than the yellow, there being only 57 % of positive responses in this light, a decrease of 8 % from that in yellow. Red light on the skin, therefore, is more effective than darkness in the production of phototropic responses, though not much more so. The nature of the reaction was the same as when the light was received through both the eye and the skin, except that the turning of the toad was followed in most cases by crawling, rather than by hopping, though the latter method of locomotion was by no means unusual.

By way of summarizing the results of the experiments with only the skin as the receptor, it may be stated that they were in all essential respects the same as when both the eye and the skin acted as receptors, and as when the eye only acted as the receptor. Blue light was the most effective stimulus, but green was not much less effective than blue, and yellow than green, the decrease in effectiveness for the two pairs of lights being the same. The lights thus formed a decreasing series with blue as the most effective stimulus, and red as the least, this light being not much more effective than darkness.

D. Reactions of Nornal Toads, compared with those of Toads from which the Eyes had been removed.

In describing the method of experimentation used in exposing only the eye to the light (p. 268), it was stated that the eye and a very small area of skin around it were illuminated. It therefore seemed desirable to ascertain whether this small area of skin had any influence upon the reactions of the toads. To test this, it was decided to make use of only the blue light, since this had proved more effective than the other lights, and if no results were obtained with it to show that this small area of skin around the eye had any influence, it was reasonable to suppose that with the other lights none would be obtained.

For these tests three toads that had not previously been experimented with were selected, and their reactions tested both in the normal condition, and after the eyes had been removed. Each toad was tested, first with the whole body, then with only the eye, and finally with only the skin exposed. The eyes were then removed and the same tests repeated. The methods and procedure were the same as have been previously described for each condition of exposure, except that a period of fifteen minutes, instead of five, was given each toad in which to react. No toad was experimented with for three days after the operation of removing the eyes. A toad from which the eyes had been excised very seldom hopped after turning, but usually crawled toward, or away from, the light.

The results of the tests are given in Table 4. Each toad was given a total of sixty-four trials, but not more than eight consecutively, an interval of at least an hour intervening before it was again tested. The total number of trials for each of the three conditions of exposure was, therefore, 192. It will be seen, by referring to Table 4, that the reactions of the toads in normal condition agreed closely with those shown in a later Table (6), also for blue light, though there were 2 % more positive responses when only the eye was exposed, than when the whole body was exposed. This, however, is not an important departure, and would probably not be present if a larger number of toads had been used. The eyeless toads, when the whole body, or only the skin, was exposed, also showed a close agreement with the

TABLE 4.

Comparison of the reactions of normal toads to blue light received through both the eye and skin, through the eye only, and through the skin only, with those of eyeless toads under the same conditions.

Condition of toads			Normal				Eyeless			
	Directions of		+	—	±	O	+	—	±	O
Reactions	Number of responses	Eye and skin	167	25			147	39	6	
		Eye region only	170	22			13	14	75	90
		Skin only	152	40			140	43	7	2
	Percent of responses	Eye and skin	87	13			77	20	3	
		Eye region only	89	11			7	7	39	47
		Skin only	79	21			73	22	4	1

The numbers under + indicate total numbers of reactions toward the light; under —, away from the light; under ±, without reference to the light (indifferent); under O, no reaction within fifteen minutes.

results obtained with hooded normal toads (Table 3), though it will be noted that in the former case there were 3 % of indifferent reactions. These results show that the hoods were an effective method of protecting the eyes from the light. But when only the region of the eye was exposed to the light, a large percentage of no reactions was obtained, with almost as large a percentage of indifferent reactions. The 7 % of positive and negative reactions were probably accidental turnings. There can be no doubt, therefore, that the method employed for exposing only the eye was an effective one, and that the reactions obtained were due to the effect of the light on the eye, and not in any way to the illumination of the small area of skin around it.

Since this narrow beam of light (blue) showed no power to stimulate the region of the eye after the eye itself had been removed, it was thought desirable to test the reactions of eyeless toads when this beam of light was thrown upon certain regions of the skin. Three regions were selected, and these may be roughly described as the region of the fore-leg, the region of the hind-leg, and the region of the back.

The results of the tests are shown in Table 5. In these tests the three eyeless toads used for the experiments just described were again employed. Each toad was given 64 trials, the total number of trials for each region being therefore 192. A period of fifteen minutes was again given each toad in which to react. It will be seen, by referring to Table 5, that positive responses were obtained when each of the three regions described was exposed to the light; but that the percentages of indifferent reactions, and of no reactions, were high, as compared with the results obtained when only the skin was exposed. This, of course, reduced considerably the percentage of positive responses which, as the Table shows, was not much above fifty, the percentage of negative responses being almost identical in the three cases. There was no evidence to show that one region was more sensitive to this form of stimulation than the others, which agrees with the results obtained by Pearse (:10) for white light. Payne (:07) has also obtained similar results on Amblyopsis. Parker (:05), however, found the tail of Ammocoetes to be the most sensitive region of the skin, and both Reese (:06) and Pearse (:10) found the same for Cryptobranchus. Reese, however, also noted that the head of Necturus was more sensitive than the tail, due probably to stimulation received through the eyes. Pearse (:10) showed that, when the eyes of Necturus were removed, the tail was the most sensitive region.

TABLE 5.

The reactions of eyeless toads to local skin illumination by blue light.

	Regions illuminated	Fore-leg				Back				Hind-leg			
Reactions	Directions of	+	—	±	O	+	—	±	O	+	—	±	O
	Number of responses	105	45	17	25	104	45	27	16	105	43	24	20
	Percent of responses	55	23	9	13	54	23	14	9	55	22	12	11

The numbers under + indicate total numbers of reactions toward the light; under —, away from the light; under ±, without reference to the light (indifferent); under O, no reaction within fifteen minutes.

The results of these experiments may now be briefly stated as follows: the reactions of eyeless toads were similar to those of hooded normal toads. When, after excision of the eye, the region of the eye was illuminated, the toads showed a large and very nearly equal percentage of indifferent reactions and failure to react. The illumination of the small area of skin around the eye had, therefore, no significant influence upon the reactions when in normal animals the eye alone was exposed to the light. The illumination of small areas of skin showed, however, that the skin could be stimulated by a narrow beam of light, similar to that used for stimulating only the eye, and that this sensitiveness was approximately the same on each of the three regions of the skin tested.

E. Summary.

The results obtained when in normal toads both the eye and the skin, the eye only, and the skin only, were exposed to the lights, as given in Tables 1–3 are repeated in Table 6. In Figure 2, are plotted the positive responses to the different lights. These results may be briefly stated as follows:

TABLE 6.

Comparison of the reactions of toads to monochromatic light received through both eye and skin, through the eye only, and through the skin only (Tables 1–3).

Monochromatic lights of equal intensity		Blue 420–480 $\mu\mu$				Green 490–550 $\mu\mu$				Yellow 570–620 $\mu\mu$				Red 630–655 $\mu\mu$			
Reactions	Directions of	+	—	±	O	+	—	±	O	+	—	±	O	+	—	±	O
Number of responses	Eye and skin	251	37			230	58			194	94			167	121		
	Eye only	246	42			214	74			187	101			160	126		2
	Skin only	296	88			271	113			248	136			217	166	1	
Percent of responses	Eye and skin	87	13			80	20			67	33			58	42		
	Eye only	85	15			74	26			65	35			55	44		1
	Skin only	77	23			71	29			65	35			57	43		

The numbers under + indicate total numbers of reactions toward the light; under —, away from the light; under ±, without reference to the light (indifferent); under O, no reaction within five minutes.

1. All the lights used produced positive responses.

2. Blue light was the most effective stimulus in the production of these responses, and green, yellow and red lights formed a decreasing series, corresponding roughly to their relative positions in the spectrum, the red being but slightly more effective than darkness.

3. The same sort of reactions were obtained when only the eye, or only the skin, was exposed, as when the whole body was exposed.

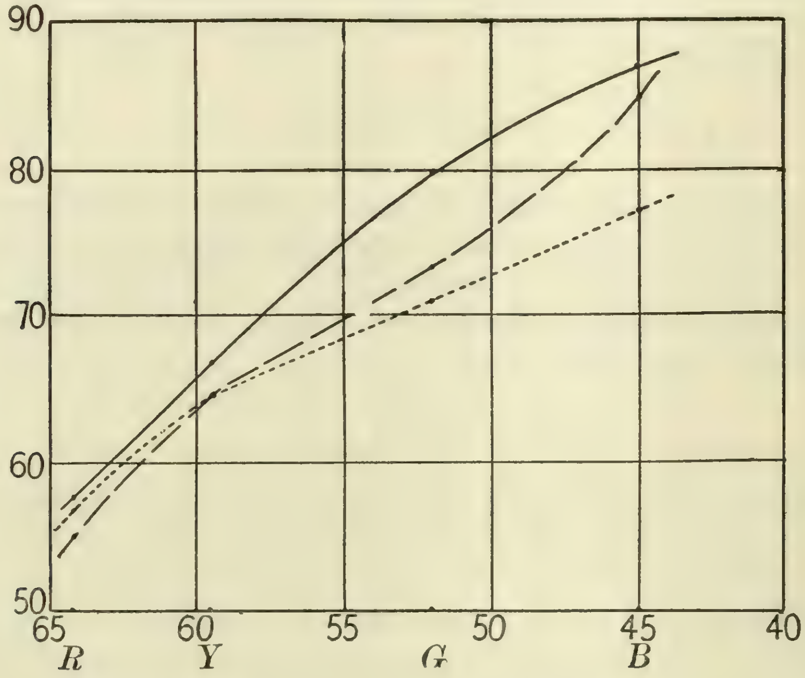

Fig. 2. Curves representing the relative distribution of effectiveness in the spectrum, when the lights are received through both the eye and the skin (————); through the eye only (- - - -); and through the skin only (. . . . . .). Wave-lengths as abscissae, and percentages of positive responses as ordinates. Points marked on axis of abscissae are the positions of the wave-lengths of the middle band of each light. *B* = Blue; *G* = Green; *R* = Red; *Y* = Yellow.

4. After the eye was excised (Table 4), exposing the eye region to a narrow beam of light produced practically no positive responses.

5. Illumination of small areas of skin by a narrow beam of light (Table 5) produced positive responses, and the same percentage of responses on each of the three areas stimulated.

2. REACTIONS TO BALANCED PAIRS OF MONOCHROMATIC LIGHTS.

A. Lights of Different Wave-lengths.

a. Reactions with both the Skin and the Eyes as Receptors.

In testing the reactions of toads to balanced pairs of monochromatic lights, the combined generators as described on p. 262–263 were used. These generators were so adjusted as to deliver lights of different wave-lengths, which entered the dark-chamber from opposite sides, the toad being placed midway between them, with the lights parallel in direction, but impinging on opposite sides of its body. The lights employed were the same as those used in the tests of the reactions to single monochromatic lights, and are described on p. 259.

The procedure, method of orientation, etc., were the same as described on p. 264–266, except that, since the lights from both generators were always used at the same time, after the first four trials had been made, the direction of rotation of the toads was simply reversed from clockwise to counter-clockwise. The orientation of each toad was therefore exactly similar to that of every other toad. A period of five minutes was again allowed each toad in which to react. The nature of the reactions was in all cases the same as that described on p. 265, except that, after the toad had turned, instead of heading toward or away from the light, it headed toward one or the other of the two lights. It might be mentioned that the reactions were usually not quite so quick as was found to be the case with the single lights.

The results of the tests are given in Table 7. The pairs of colored lights were arranged in order, according to their distribution in the spectrum. There are thus seen to be three groups of pairs of lights. The first group, represented by one pair, was made up of the two lights farthest apart in the spectrum, the blue and the red, the wave-lengths of their middle bands differing by 192.5 $\mu\mu$. In the second group, represented by two pairs, the lights in each pair were nearer each other in the spectrum than were the blue and the red. These pairs were the blue and yellow, and the green and red, the wave-lengths of the middle bands of the lights of each pair differing by 145.0 $\mu\mu$ and 122.5 $\mu\mu$, respectively. The third group was represented by three pairs of lights, each pair being composed of the lights that are adjacent to each other in the spectrum, viz., the green and yellow, the blue and green, and the yellow and red, the differences of the wave-lengths

TABLE 7.

Reactions of toads to balanced lights of different wave-lengths received through both skin and eyes.

		Group 1.				Group 2.								Group 3.											
Pairs of balanced colored lights		Blue and Red 420–480 μμ 630–655 μμ				Blue and Yellow 420–480 μμ 570–620 μμ				Green and Red 490–550 μμ 630–655 μμ				Green and Yellow 490–550 μμ 570–620 μμ				Blue and Green 420–480 μμ 490–550 μμ				Yellow and Red 570–620 μμ 630–655 μμ			
Distances in wave-lengths between the middle bands of pairs of lights		192.5 μμ				145.0 μμ				122.5 μμ				75.0 μμ				70.0 μμ				47.5 μμ			
Reactions	Directions of	B	R	±	O	B	Y	±	O	G	R	±	O	G	Y	±	O	B	G	±	O	Y	R	±	O
	Number of responses	788	167	5		678	278	3	1	718	236	6		651	293	16		592	365	1	2	632	323	2	3
	Percent of responses	82	17	1		71	29			75	24	1		68	30	2		62	38			66	34		

The numbers under *B* indicate total numbers of reactions toward the blue; under *G*, toward the green; under *R*, toward the red; under *Y*, toward the yellow; under ±, without reference to either color (indifferent); under O, no reaction within five minutes.

of the middle bands among these being 75.0 μμ, 70.0 μμ, and 47.5 μμ, respectively.

Five sets of twelve toads each were tested in each of the six double pairs of lights, and since each toad was given eight trials in each combination of lights, the total number of trials for the five sets was 960 in each single pair of lights or 5760 trials in all. Three of the five sets of twelve toads were the same as were used in testing the reactions to single monochromatic lights, when both the eye and the skin acted as receptors. Two new lots of twelve toads each were, however selected for the other two sets. These tests were, therefore, carried out on 36 separate toads.

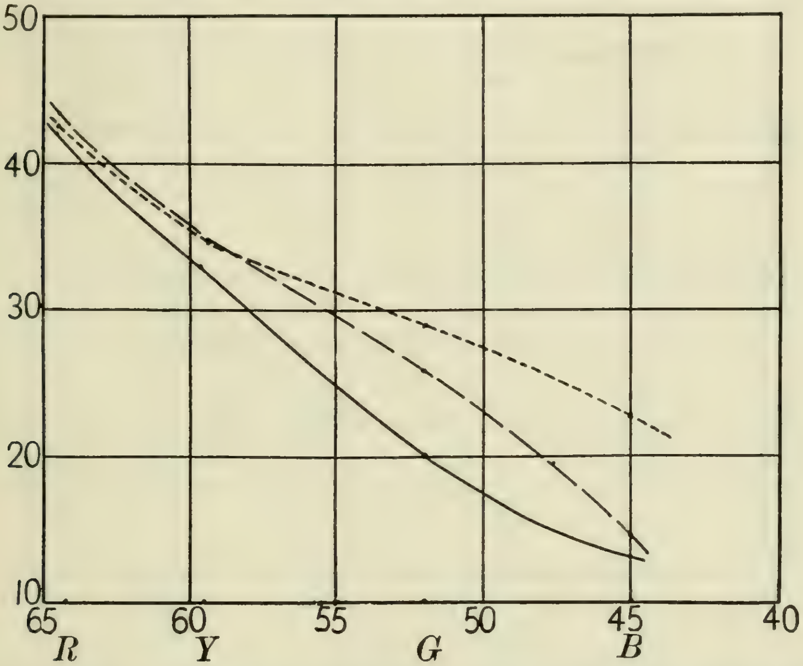

FIG. 3. Curves representing the percentages of negative responses to the four sets of wave-lengths, when received through both the eye and the skin (————); through the eye only (- - - -); and through the skin only (. . . . . .). Wave-lengths as abscissae and precentages of negative responses as ordinates. Points marked on axis of abscissae are the positions of the wave-lengths of the middle band of each light. *B* = Blue; *G* = Green; *R* = Red; *Y* = Yellow.

It will be seen, by referring to Table 7, that the results of the tests of the reactions to balanced pairs of monochromatic lights were, in the main, very similar to those obtained in response to single monochromatic lights under the same conditions. The numbers under each pair of lights represent the combined results of two sets of trials, those in which the light with the shorter wave-lengths in each pair proceeded from generator *A*, and those in which it proceeded from generator *B*. In this way it was hoped to eliminate possible errors in the working of the generators. However, though the results were, in the main, the same as those obtained with single monochromatic lights, it cannot be said that there is much evidence in favor of positive phototropism for the red light. There were, for each pair of lights, some movements toward both lights, with the larger percentage always to the blue, or to that light of a given pair which in the spectrum was nearer the blue. Even in the pairs in which red occurred, there were movements toward this light. But, when the lights with which red was paired were used singly, there were movements to the dark, that is, movements away from these lights. If the results obtained here are compared with those given in Table 1, and the curves shown in Fig. 3 compared with those shown in Fig. 4, it will be seen that the percentages of movements toward the red, when paired

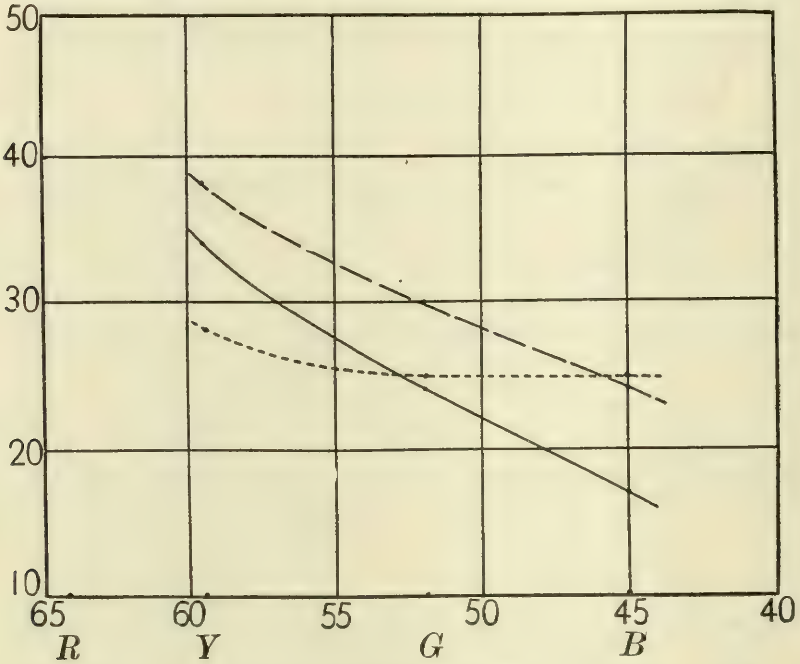

FIG. 4.— Curves representing the percentages of movements toward red when paired with the three other lights, when the lights are received through both the skin and the eyes (————); through the eyes only (- - - -); and through the skin only (. . . . . .). Wave-lengths as abscissae and percentages of movements as ordinates. Points marked on axis of abscissae are the positions of the wave-lengths of the middle band of each light. *B* = Blue; *G* = Green; *R* = Red; *Y* = Yellow.

with other lights, were only a little higher than those toward the dark when these lights were used singly; in other words, the negative responses to blue, green, and yellow, when paired with red light, were almost the same as when these lights were used singly. Red light can be said, therefore, to have had only very little, if any, effect in the production of positive responses, when used in pairs with other lights, while blue, green, and yellow were distinctly effective in this respect.

Although for each pair of lights, there were always positive responses to the blue, or to the light which in the spectrum was nearer the blue, still the distribution of effectiveness, as seen in the percentage of responses to the light of shorter wave-lengths in any pair, did not follow closely the distribution of the several lights in the spectrum. Nevertheless, when the blue light was paired in sequence with the other three lights, it was found that the percentage of positive responses

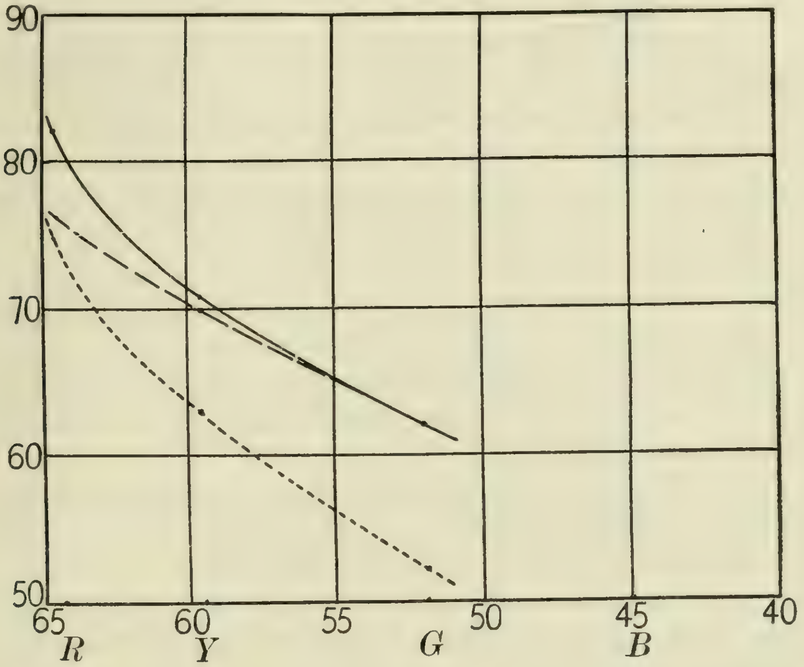

Fig. 5. Curves representing the percentages of movements toward blue when paired with the three other lights, when the lights are received through both the skin and the eyes (———); through the eyes only (- - - -); and through the skin only (. . . . . .). Wave-lengths as abscissae and percentages of movements as ordinates. Points marked on axis of abscissae are the positions of the wave-lengths of the middle band of each light. *B* = Blue; *G* = Green; *R* = Red; *Y* = Yellow.

to the blue decreased as the spectral distance of the other light from the blue decreased (see also Fig. 5). And if the same were done for the red, it was found that the percentage of positive responses to the other three lights increased as the distance from the red increased (see also Fig. 4).

Although the blue was considerably more effective than the green when these two lights were paired, the percentage of responses being in the ratio of 62 to 38, yet, when these two lights were paired with yellow, there was only 3 % more responses to blue than to green (see Table 7, and Fig. 6); and again, when they were paired with red (see Table 7, and Fig. 4), there was 7 % more responses to blue than to green. By referring to Table 1, it will be seen that there was a difference of 7 % between the effectiveness of the blue and green when these lights were used singly. Green was also considerably more effective than yellow when paired with it, the ratio of the percentage of responses being 68 to 30, with two indifferent reactions. But, when green and yellow were paired consecutively with blue (see Table 7, and Fig. 5), the greater effectiveness of green over yellow was much lessened, and there was only 9 % more responses to the green than to the yellow. When these two lights were similarly paired with red (see Table 7, and Fig. 4), the difference in effectiveness was again found to be only 9 %. There was a difference of 13 % between the effectiveness of these two lights when used singly in the production of positive responses (Table 1).

By way of summarizing the results of the experiments with balanced pairs of monochromatic lights in which both the skin and the eyes acted as receptors, it may be said that they were in general similar to those obtained with single monochromatic lights under the same conditions. Blue, green, and yellow were effective, in the order given, in the production of positive responses; red produced only a very slight positive response, if any at all. There were movements toward both lights in any pair, with the larger percentage always toward the light nearer the blue end of the spectrum. Blue was the most effective stimulus in the production of positive responses, green next, and yellow next, while red light had little, or no more effect than darkness.

b. Reactions with the Eyes as Receptors.

While the results of the tests of the reactions to single monochromatic lights, in which only the eye, and only the skin, were exposed to the light showed the same general results as those obtained when the whole body was exposed, there were a few points of difference in the reactions to each. It therefore seemed worth while to test the reactions to balanced pairs of monochromatic lights, exposing first only the eyes; and secondly, only the skin. In testing the reactions in which only the eyes served as receptors, the method and procedure followed were the same as those described on p. 264–266, with the modifications given on p. 268 and 277.

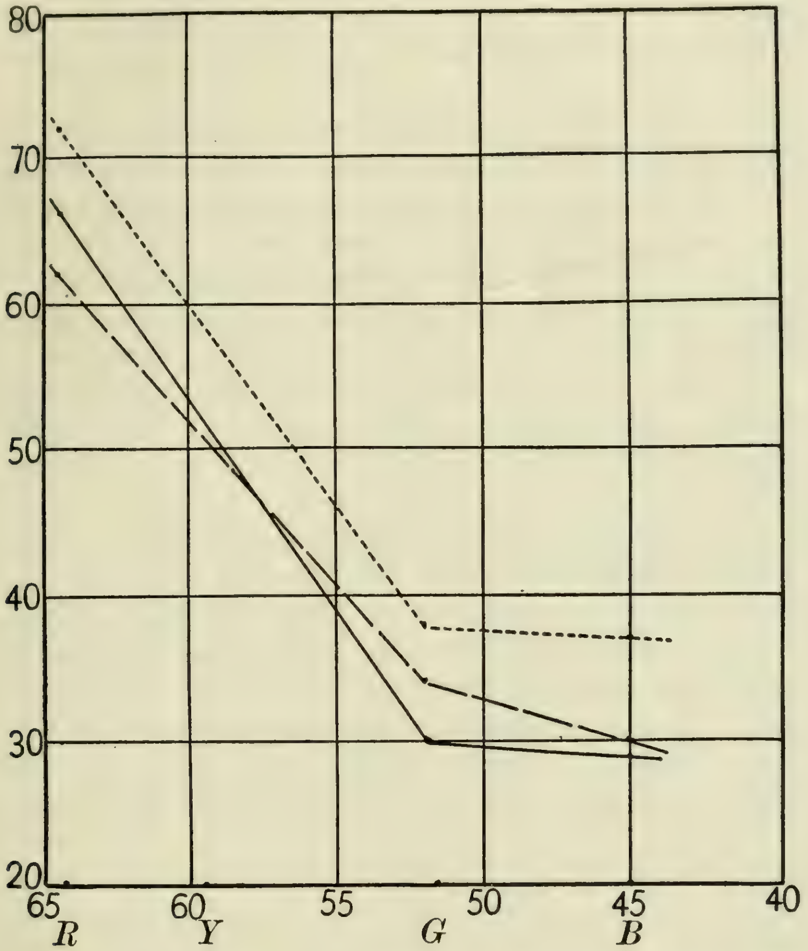

FIG. 6.— Curves representing the percentages of movements toward yellow when paired with the three other lights, when the lights are received through both the skin and the eyes (———); through the eyes only (- - - -); and through the skin only (. . . . .). Wave-lengths as abscissae and percentages of movements as ordinates. Points marked on axis of abscissae are the positions of the wave-lengths of the middle band of each light. *B* = Blue; *G* = Green; *R* = Red; *Y* = Yellow.

The results of the tests are given in Table 8. Two sets of twelve toads each were tested, making a total of 384 trials in each single pair of lights or 2304 trials in all. One of the sets of twelve toads were the remainder of the lot used for the first three sets of reactions, when both the eyes and skin acted as receptors, though seven individuals died during the course of the experiments, and their places had to be supplied by seven new toads. The other lot employed was one of the new lots selected for the last two sets of tests in the same series of experiments.

By referring to Table 8, it will be seen that the results were, in general, the same as those obtained when the whole body was exposed (Table 7). In any pair of lights there were movements toward both lights, with the larger percentage of responses to the blue, or to the light of a given pair which, in the spectrum, is nearer the blue. There were only a few more movements to red light, however, in the pairs with green and yellow, than there were negative responses to these lights, when they were used singly; though, when paired with blue, there was 9 % more (Table 2, and Figs. 3 and 4). Red light under these circumstances was, therefore, not much more effective than darkness in the production of responses.

When blue and green were paired, the percentage of responses to the blue was considerably higher than that to the green, the ratio being 62 to 38. But, when blue or green was paired with yellow (see also Fig. 6), there was only 4 % more responses to blue than to green; and when paired with red (see also fig. 4), only 6 % more. There was 11 % more responses to blue than to green when they were used singly (Table 2). Green also was considerably more effective than yellow when these two lights were paired, the percentage of responses to them being in the ratio of 66 to 34. But when paired with blue consecutively (see also Fig. 5), the difference in effectiveness was only 8 %, and when paired with red (see also Fig. 4), also only 8 %. The difference in effectiveness between green and yellow, when used singly, was 9 % (Table 2).

By way of summarizing the results of the experiments with balanced pairs of monochromatic lights in which only the eyes served as receptors, it may be said that they were essentially the same as when both the skin and the eyes acted as receptors, and in the main, the same as those obtained with single monochromatic lights, when only the eye was exposed. Blue, green, and yellow were effective in the production of positive responses, but red, in comparison with any other color, never showed this. Blue was the most effective stimulus, and red the least, and the decrease in effectiveness followed the order of the

TABLE 8.

Reactions of toads to balanced lights of different wave-lengths received through the eyes only.

	Group 1.				Group 2.								Group 3.											
Pairs of balanced colored lights	Blue and Red 420–480 μμ 630–655 μμ				Blue and Yellow 420–480 μμ 570–620 μμ				Green and Red 490–550 μμ 630–655 μμ				Green and Yellow 490–550 μμ 570–620 μμ				Blue and Green 420–480 μμ 490–550 μμ				Yellow and Red 570–620 μμ 630–655 μμ			
Distance in wave-lengths between the middle bands of pairs of lights	192.5 μμ				145.0 μμ				122.5 μμ				75.0 μμ				70.0 μμ				47.5 μμ			
Reactions: Directions of	B	R	±	O	B	Y	±	O	G	R	±	O	G	Y	±	O	B	G	±	O	Y	R	±	O
Number of responses	290	94			270	114			267	117			252	132			239	145			237	147		
Per cent of responses	76	24			70	30			70	30			66	34			62	38			62	38		

The numbers under *B* indicate total numbers of reactions toward the blue; under *G*, toward the green; under *R*, toward the red; under *Y* toward the yellow; under ±, without reference to either color (indifferent); under O, no reaction within five minutes.

colors in the spectrum. There were movements toward both lights in any pair, the larger percentage being always to the blue, or to the light nearer the blue end of the spectrum. The percentage of movements to red light in pairs with the other lights, was only slightly higher than the percentage of movements toward the dark, when these lights were used singly.

c. Reactions with the Skin as Receptor.

Having demonstrated that the eyes were concerned in the reactions in which the whole body was exposed, it remained to test the reactions of toads in which only the skin was exposed to balanced pairs of monochromatic lights. In order to protect the eyes from the light, the hoods described on p. 269 were again used.

The results of the tests are given in Table 9. Seven sets of twelve toads each were tested, and therefore a total of 1344 trials was made in each single pair of lights, or 8064 trials in all. For the first three sets, four of the lot of twelve toads which had been employed for the first three sets of tests where both the skin and the eyes were exposed, and for the first set of tests where only the eyes were exposed, were used. Eight of this lot of twelve had died during the course of former experiments, and their places were therefore supplied by eight other toads. Three of the present lot of twelve also died, during the course of these experiments, and their places were supplied by three other animals. For the next two sets, a new lot of twelve, and for the last set, another new lot of twelve toads, were again selected. One of each of these last two lots also died during the course of these experiments, and their places were supplied by two other animals. There were thus a total of 41 separate toads used for these tests.

It will be seen, by referring to Table 9, that in all of the pairs there were some movements toward both lights, with the larger percentage always toward the blue, or to that light of a given pair which, in the spectrum, is nearer the blue. In the pairs in which red occurred, there were also movements to this light, but only when paired with blue were these movements to red light more numerous than the negative responses to the lights when used singly. It was here, in these pairs of red with the other lights, that were most clearly brought out the differences in sensitiveness, when the light was received through the eyes, or through both the skin and the eyes, and that when it was received through the skin only. If the pairs in which red occurred be considered, it will be seen that the percentage of movements

TABLE 9.

Reactions of toads to balanced lights of different wave-lengths received through the skin only.

		Group 1.				Group 2.								Group 3.											
	Pairs of balanced colored lights	Blue and Red 420–480 $\mu\mu$ 630–655 $\mu\mu$				Blue and Yellow 420–480 $\mu\mu$ 570–620 $\mu\mu$				Green and Red 490–550 $\mu\mu$ 630–655 $\mu\mu$				Green and Yellow 490–550 $\mu\mu$ 570–620 $\mu\mu$				Blue and Green 420–480 $\mu\mu$ 490–550 $\mu\mu$				Yellow and Red 570–620 $\mu\mu$ 630–655 $\mu\mu$			
	Distances in wave-lengths between the middle bands of pairs of lights	192.5 $\mu\mu$				145.0 $\mu\mu$				122.5 $\mu\mu$				75.0 $\mu\mu$				70.0 $\mu\mu$				47.5 $\mu\mu$			
Reactions	Directions of	B	R	±	O	B	Y	±	O	G	R	±	O	G	Y	±	O	B	G	±	O	Y	R	±	O
	Number of responses	1005	339			841	500	3		1004	338	2		830	513	1		702	642			968	374	2	
	Percent of responses	75	25			63	37			75	25			62	38			52	48			72	28		

The numbers under *B* indicate total numbers of reactions toward the blue; under *G*, toward the green; under *R*, toward the red; under *Y*, toward the yellow; under ±, without reference to either color (indifferent); under O, no reaction within five minutes.

toward the lights with which it was paired, was the same for the blue and for the green, and only 3 % less for the yellow (see also Fig. 4). This did not point, as might be supposed, to the ineffectiveness of the red light on the skin alone when paired with others, for that had already been shown to be the case not only for the eyes and the skin when both were exposed to the light, but also for the eyes alone; it did show, however, that the sensitiveness of the skin to differences in wave-lengths, at the blue end, when it was exposed to balanced pairs of monochromatic lights, was much lower than that of the eyes, or of the eyes and the skin together. This fact was further brought out when yellow was paired with blue and green. There was only 1 % more of responses to blue than to green when paired with yellow (see also Fig. 6), and only 4 % more to blue than to green when these lights were directly paired, the percentage of responses to each being in the ratio of 52 to 48. When blue and green were used singly, they were also near in their effect on the skin (Table 3).

The skin, however, did show some differences in sensitiveness to lights of different wave-lengths. If the pairs of lights in which blue occurred be considered, it will be seen that the percentage of responses to the blue decreased as the spectral distance of the other light from the blue decreased, until, in the pair blue and green, the movements to each light were practically the same, there being only a difference of 4 % between them (see also Fig. 5). Green when paired with yellow, however, showed a considerably greater effectiveness, the ratio of the percentage of responses being 62 to 38; also when these two lights were paired with the blue (see also Fig. 5), there was a difference of 11 % between them, though, when they were paired with red (see also Fig. 4), there was a difference of only 3 % between them, which was, of course, due in greater part to the ineffectiveness of the red light.

By way of summarizing the results of the experiments with balanced pairs of monochromatic lights in which only the skin acted as a receptor, it may be stated that the results do not correspond entirely with those obtained when either the whole body, or only the eyes, were exposed to balanced pairs of monochromatic lights, or when the skin only was exposed to single monochromatic lights. While blue, green, and yellow were effective in the order in which they are given in the production of responses, the difference in effectiveness between blue and green was hardly noticeable, though that between green and yellow was considerable. Red, again, shows itself to be but little more effective than darkness. The sensitiveness of the skin to lights of different wave-lengths, while much reduced at the most effective end of the spectrum, still must be regarded, in all essential respects, as similar to that of the eyes, though much reduced throughout.

B. Lights of the same Wave-lengths.

Reactions with both the Skin and the Eyes as Receptors.

There could hardly be any doubt that the lights from the two generators were the same, not only in the range of wave-lengths shown to occur in them by spectroscopic examination, but also in their relative intensities, as shown by radiomicrometric readings. It seemed desirable, however, to test further these conditions experimentally, and the following trials were accordingly made.

The generators were so adjusted as to deliver lights of the same wave-lengths and intensities, and the toads were tested in the field between these two presumably equal sources. The results of these tests are given in Table 10. Two sets of twelve toads each were tested, each set being given 96 trials in each pair of lights, and therefore a total of 192 trials for the two sets of toads. The procedure, orientation, etc., were precisely the same as carried out in the tests of the

TABLE 10.

Reactions of toads to balanced lights of the same wave-lengths received through both skin and eyes.

Pairs of balanced colored lights		Blue and Blue				Green and Green				Yellow and Yellow				Red and Red			
Reactions	Directions of	A	B	I	O	A	B	I	O	A	B	I	O	A	B	I	O
	Number of responses	92	97	3		94	90	8		90	87	15		78	83	31	
	Percent of responses	48	50	2		49	47	4		47	45	8		41	43	16	

The numbers under *A* indicate total numbers of reactions toward the light from generator *A*; under *B*, toward the light from generator *B*; under *I*, without reference to either light (indifferent); under *O*, no reaction within five minutes.

reactions to balanced pairs of lights of different wave-lengths. By referring to Table 10, it will be seen that the two generators *A* and *B* delivered lights that were very similar in effect, the differences in the percentages of reactions to one or the other of a given pair being hardly noticeable, and not constant for either. The decrease in the percentage of positive responses between blue and red shown here was very slight, there being 98 % of positive responses in the blue, 96 % in the green, 92 % in the yellow, and 84 % in the red, the decrease between the yellow and red being the greatest of that between any pair of lights adjacent to each other in the spectrum. This decrease from blue to red was, however, sufficient to bring out very clearly a gradually increasing percentage of indifferent reactions, there being about eight times as many indifferent reactions in red as in blue.

These results served to confirm the belief that the spectroscopic and photometric readings of the different lights had been sufficiently accurate as a means of keeping constant the radiant energy contained in each light.

By way of summarizing these results it may be said that the two generators gave out lights that were very similar in effect, and that therefore the combining of the results of two sets of trials in which the same lights occurred, but from different generators, was a legitimate procedure.

C. Summary.

The results obtained with balanced pairs of monochromatic lights are summarized in Table 11. These may be briefly stated as follows:

1. The results obtained with balanced pairs of monochromatic lights agree in all essential respects with those obtained with single monochromatic lights.

2. Blue, green, and yellow lights produced positive responses of a marked degree; red light resembled darkness, or called forth only a very slight positive response.

3. Blue light was the most effective stimulus; and green, yellow, and red formed a series of decreasing effectiveness, corresponding roughly to their distribution in the spectrum.

4. In any pair of balanced lights the larger percentage of responses was toward the light which in the spectrum was nearer the blue end.

5. In effectiveness the light nearer the blue end of the spectrum, in the several pairs of lights tried, did not correspond very closely to that of the pairs of lights as determined by their distances apart in the spectrum.

TABLE 11.

Comparison of the reactions of toads to balanced lights of different wave-lengths received through both skin and eyes, through the eyes only, and through the skin only (Tables 7–9).

	Group 1.				Group 2.								Group 3.											
Pairs of balanced colored lights	Blue and Red 420–480 μμ 630–655 μμ				Blue and Yellow 420–480 μμ 570–620 μμ				Green and Red 490–550 μμ 630–655 μμ				Green and Yellow 490–550 μμ 570–620 μμ				Blue and Green 420–480 μμ 490–550 μμ				Yellow and Red 570–620 μμ 630–655 μμ			
Distances in wave-lengths between the middle bands of pairs of lights	192.5 μμ				145.0 μμ				122.5 μμ				75.0 μμ				70.0 μμ				47.5 μμ			
Reactions — Directions of	B	R	±	O	B	Y	±	O	G	R	±	O	G	Y	±	O	B	G	±	O	Y	R	±	O
Number of responses — Eyes and skin	788	167	5		678	278	3	1	718	236	6		651	293	16		592	365	1	2	632	323	2	3
Number of responses — Eyes only	290	94			270	114			267	117			252	132			239	145			237	147		
Number of responses — Skin only	1005	339			841	500	3		1004	338	2		830	513	1		702	642			968	374	2	
Percent of responses — Eyes and skin	82	17	1		71	29			75	24	1		68	30	2		62	38			66	34		
Percent of responses — Eyes only	76	24			70	30			70	30			66	34			62	38			62	38		
Percent of responses — Skin only	75	25			63	37			75	25			62	38			52	48			72	28		

The numbers under *B* indicate total numbers of reactions toward the blue; under *G*, toward the green; under *R*, toward the red; under *Y*, toward the yellow; under ±, without reference to either color (indifferent); under O, no reaction within five minutes.

6. The same sort of reactions were obtained when only the eyes were exposed, as when the whole body was exposed, but there was a considerable decrease in the sensitiveness of the skin to differences in wave-lengths, compared with that of the eyes, or of the eyes and the skin.

IV. Discussion.

Sensitiveness to differences in wave-lengths is evidently a quality residing, not only in the eyes of the toad, but in the skin as well. It has been shown that the reactions when the skin alone was exposed to the lights were essentially the same as those when the eyes, or when the eyes and the skin, were exposed. It is necessary, therefore, to consider somewhat the nature of the photoreceptors in the eyes and in the skin. In the eye there is the retina, with its rod- and cone-cells, i. e., nerve-terminations differentiated for the reception of ether waves, which set up chemical changes in the rods and cones, and thus give rise to nerve impulses that are transmitted to the brain, and there perceived as light. In the skin are found the terminations of the spinal nerves. We are able to form some idea of how different lights appear to the toad when received through the eyes, but when we attempt to consider how they would appear when they are received through the skin alone, the problem becomes much more difficult. It has been found that in some amphibians there is a connection between the nerve terminations of the eyes and those of the skin, as is to be inferred from results when the skin is illuminated. Engelmann ('85) found that changes took place in the retinae of frogs when only the skin was exposed to light; though Fick ('90) obtained results which led him to conclude that interference with the normal respiration was the cause of these changes. Koranyi ('92) noted that illumination of the skin of the frog caused microscopic changes in the retina similar to those produced by the illumination of the eye itself.

It is generally believed by modern physiologists that the perception of color is a function of the cones alone, and that the rods are sensitive only to light and darkness; and that, by virtue of their power of adaptation in the dark, through the regeneration of visual purple, they form the special apparatus for vision in dim light. The generally accepted theories of color-vision all presuppose the existence of photochemical substances in the eye, which, when acted upon by the different wave-lengths of the visible spectrum, undergo different chemical changes, which give rise to different nerve-impulses that are transmitted to the brain, and there call forth different color sensations. Whether these photo-chemical substances exist as well in the skin, it is impossible to say, but, in so far as the reactions of toads are concerned, the specific chemical effects of the different wave-lengths upon the nerve terminations in the skin seem to be similar to those upon the nerve terminations in the retina; i. e., the nerve-impulses which are set up by the specific chemical changes due to the effects of the several lights, seem to be similar in the two sets of terminations. Parker (:**03**, p. 33) expressed the view that "the positive phototropism of eyeless frogs depends upon the capacity of the nervous structures of the frog's skin to be stimulated by light." This capacity for stimulation in the nervous terminals of the skin, which seems also to be present in many other amphibians, including the toad (Pearse, :**10**), must now be extended, as the present work shows, to lights of different wave-lengths.

The results of the tests of reactions to single monochromatic lights received through the eye, through the skin, and through both organs, have been given together in Table 6. The results in these three cases were substantially the same, with only a few minor points of difference. All the lights produced positive responses in each of the three cases, and the sequence of the lights, as determined by their effectiveness in stimulating the toads, corresponded to their sequence in the spectrum, with blue as the most effective stimulus, and red as the least. Although the decrease in the effectiveness of the four lights to call forth positive responses follows the order of the spectrum from blue to red, the distribution of this effectiveness does not correspond very closely to the distribution of the several lights in the spectrum. If each light be designated by the wave-length of its middle band, the position of the four lights in the spectrum would be as follows: blue, 450 $\mu\mu$; green, 520 $\mu\mu$; yellow, 595 $\mu\mu$; and red, 642.5 $\mu\mu$. The respective distances are therefore: between the blue and green, 70.0 $\mu\mu$; between the green and yellow, 75.0 $\mu\mu$; and between the yellow and red, 47.5 $\mu\mu$. It is seen that the yellow and red were the nearest of any pair in the spectrum, but in no one of the conditions of exposure to the light, was this pair of lights the nearest in effect in the production of positive responses. In the reactions in which the eye and skin were both exposed the blue and green were the nearest in effect in the production of positive responses; in those in which only the eye was exposed, the green and yellow were the nearest; and in those in which only the skin was exposed, the difference in effectiveness in the production of positive responses was the same between blue and green, and between green and yellow.

By referring to Table 6 and to Figure 2, in which the positive responses to the different lights are plotted, it will be seen that the percentage of positive responses in the most effective light was higher when received through the eye alone, than when received through the skin alone. The green light had also a greater effect when received through the eye alone than when received through the skin alone, but the difference between the two receptors was very small. In the yellow light the effect on the eye and the skin was the same, while in the red, the percentage of positive responses was slightly higher for the skin than for the eye. The blue and green, therefore, differed more in effect when received through the eye alone, than when received through the skin alone. The green and yellow, and the yellow and red, also showed this greater difference in effectiveness when received through only the eye, than when received through only the skin, though the differences in effectiveness on the eye were, in these last two cases, not so much greater than those on the skin, as they were in the blue and green.

The differences in sensitiveness shown by the eye and the skin in the blue and green lights were brought out when both the eye and the skin served as receptors. The blue showed a higher percentage of effectiveness when received through both the eye and the skin, than when received through only one or the other, though this percentage of effectiveness was not much higher than that obtained when only the eye acted as a receptor. The green showed the same increase in effectiveness when both the eye and the skin served as receptors, but it was considerably higher than when only the eye acted as a receptor. Blue light, therefore, was not so much more effective than the green on the eye and the skin together, as it was when the lights were received through the eye only. This decrease in the effectiveness, of blue over green, can probably be explained as follows: the sensitiveness of the eye to blue light was much greater than to green light. The sensitiveness of the skin to blue light was considerably lower than that of the eye, but the sensitiveness of the skin to green light was not much lower than that of the eye. Therefore, when the light was received through both these receptors, the effectiveness of blue light over that of green was considerably decreased. In both the yellow and the red, the percentage of positive responses, when both the eye and the skin were exposed, was very close to that obtained when only the one or the other was exposed. Since green light was more effective when received through both the eye and the skin than when received through only the eye, there was a greater difference in effectiveness between green and yellow, when received through both the eye and the skin, than when received through only the eye.

It is clear, therefore, that, while the reactions of the toads to single monochromatic lights, when only the skin was exposed, were, in all essential respects, the same as those when only the eye, or the whole body was exposed, the most effective light — blue — was more effective on the eye than on the skin, as was also the green, but less so than the blue, while the yellow and red showed an almost equal effectiveness through the skin, and through the eye. The difference in the effectiveness of blue and green on the eye and skin was clearly shown when both the eye and the skin acted as receptors.

When we come to consider the reactions to balanced pairs of monochromatic lights, it is found that the relations of the eyes, the skin, and the whole animal, were very much the same as those found in the single lights. The relation of the distribution of effectiveness and the distribution of the several lights in the spectrum was also very similar to that in single lights. The distribution of effectiveness followed closely the distribution of the several lights in the spectrum, neither when one light was paired consecutively with the others, nor when the pairs of lights were considered according to the distance apart of the lights in each pair in the spectrum. The pairs of lights were arranged in the tables in sequence, according to the distance apart in the spectrum of the lights in each pair, beginning with the two farthest apart, and ending with the two nearest together. But in not one of the three conditions of exposure, did the distribution, according to effectiveness of the more refrangible light in each pair, correspond to the order in which the pairs of lights were placed. It came nearest to doing so when only the eyes were exposed. When only the skin was exposed, the lack of stimulating effect of the red light on the skin was, I think, clearly the chief cause of the lack of agreement between the two series of tests. But when both the eyes and the skin were exposed to the lights, though the ineffectiveness of the red light probably in great part explained the lack of agreement between the effectiveness and the distribution of the lights, still, the presence of fewer positive responses to the blue when paired with the yellow, than there were positive responses to the green when paired with the red, was also due, in some part, to the fact that, when used singly, blue light was slightly nearer to green in effect than was yellow to red; and therefore, when blue was paired with yellow, there were fewer responses to blue, than there were to green, when the latter was paired with red. The same reasoning applies to the pairs blue with green, and yellow with red, and explains why there were more positive responses to yellow than to blue in these pairs. In effect green and yellow were the farthest apart of the lights adjacent to each other in the spectrum, as was found to be the case in the reactions to the single lights.

When we proceed to the consideration of the reactions where only the eyes were exposed to the lights, it is found that the decrease in the responses to the more refrangible light in the several pairs followed pretty closely the order in which the pairs of lights were placed in the Table, that is, there was a more evenly graded series. This was, it will be remembered, also found to be the case in the reactions to single lights.

The difference in effectiveness between blue and green, when tested in single lights, was only 1 % greater than that between yellow and red; therefore, when blue was paired with yellow, and green with red, there was the same percentage of responses to the green as there was to the blue, though from the relative position in the spectrum of the lights in each of these pairs, we might have expected more responses to the blue than to the green. When blue was paired with green, and yellow with red, there was also the same percentage of responses to the yellow as to the blue. The ineffectiveness of the red light, in the pairs green and red, and yellow and red, of course also had something to do with the equality of responses to the blue and to the green in the pairs blue and yellow, and green and red; and also with the equality of responses to the blue and to the yellow in the pairs blue and green, and yellow and red, respectively.

When the lights were received through the skin only, there was brought out a greater lack of sensitiveness to differences in wave-lengths, particularly at the more refrangible end of the spectrum, than when the light was received through the eyes only. In the reactions to single lights, when received through only the skin, there was seen to be less difference in effectiveness between blue and green, green and yellow, and yellow and red, than when the eye alone acted as a receptor. But here, when exposed to balanced lights, this greater lack of sensitiveness on the part of the skin to differences in wave-lengths was seen particularly at the blue end, there being a considerable difference between the effectiveness of the yellow and the red. The green and yellow also differed markedly in their relative effectiveness, both when paired together, and when paired with blue; though when paired with red, they were very similar, owing to the ineffectiveness of the latter.

Red light, as has already been pointed out, was less effective in pairs with other lights when received through only the skin, than when received through only the eyes. When paired with blue light, there was 1 % more of movements toward red than there was when the lights were received through only the eyes. From a comparison with the results obtained with single lights, we might have expected the responses to blue light when received through only the eyes, to be higher than when received through only the skin.

As was found to be the case in the reactions to single lights, the effects of the stimulation of the two kinds of receptors showed their influence upon the reactions when the whole animal was exposed to the light. The more effective light, in most of the pairs of lights, showed a higher percentage of responses when received through both the eyes and the skin, than when received through only one of the two. The pairs of red with green and yellow, however, do not conform to this, owing to the greater effectiveness of the red light when received through only the eyes, as compared with that when it was received through only the skin. The blue and green had also the same effect on both the eyes and the skin, as on the eyes alone. This was due to the lack of sensitiveness in the skin to differences in wave-lengths at the blue end, when the light was received through only the skin. The green and red showed the same percentage of positive responses to the green when received through both the eyes and the skin, as when received through the skin alone. This was due to the ineffectiveness of red light to stimulate the skin, as well as to the less sensitiveness of the skin to differences in wave-lengths. The blue light, in pairs with other lights, had very little more stimulating value for the skin than had the green, in pairs, when only the eyes were exposed; still, the blue was plainly more potent than the green. When both the skin and the eyes were exposed to the lights, the blue was no more potent than was the green, when the lights were received through only the eyes. This was again due to the fact that the sensitiveness of the skin to differences in wave-lengths in the more refrangible lights of the spectrum was very slight, and that what differences were found in the reactions, when both the skin and the eyes were exposed, were due, for the most part, to the sensitiveness of the eyes.

There is seen in these reactions to balanced pairs of lights a counter effect of the different sets of wave-lengths. Each of the lights in a given pair seemed to be able to exert its influence on the reactions. The more potent, short-waved light, in any pair, reduced considerably the effect of the less potent, long-waved light, as measured by their percentages of positive responses when used singly. But the less effective light also reduced the effect of the more potent light. The less effective lights, yellow and red, made the two more effective — blue and green — much more nearly similar in their effects, when paired with them, than when blue and green were paired together, or used singly. When the lights were received through only the skin, this was made even more evident, owing to the comparatively greater insensitiveness of the skin to differences in wave-lengths, particularly in the more refrangible lights.

The reactions to balanced pairs of monochromatic lights showed, therefore, essentially the same relations under the three different conditions of exposure, and also the same relations as in the reactions to single lights. But the sensitiveness of the skin to differences in wave-lengths was not as great as that of the eyes. Moreover, the effectiveness of the more potent light in any pair is reduced by that of the light with which it is paired, and *vice versa*; and this has a tendency to make the differences in effect between the more effective lights less, when paired with others, than when they were used singly, or when paired with each other.

The slight lack of agreement between the distribution of the effectiveness of the lights and the distribution of the several lights in the spectrum must be due to specific chemical effects called forth by the several lights. And the fact that the distribution of the effectiveness of the several lights was different for the three conditions of exposure, points to the conclusion that the effects on the eye were slightly different from those on the skin, and that, therefore, the reactions when both the skin and the eyes served as receptors should also be different from those when one or the other served alone. The toad probably reacted, not to the stimulation of the light, directly, but to the chemical changes which were produced in the eyes and the skin by the lights. These chemical changes were greatest in the blue, and least in the red, and while the sequence of effectiveness followed the spectrum in this order, the lights between blue and red had each their own specific effects, which were different in amount, though not in kind, on the eyes and on the skin. It is not known exactly how light affects chemical reactions, or what the chemical changes are that take place upon illumination, but that they are a function of the wave-lengths has been brought out by the present experiments. The absorption of the light surely plays a part, for, if the light were not absorbed, the reactions would not have taken place. The effectiveness of the several lights, however, cannot be attributed to the energy of the light alone, for all the lights used in these experiments were very closely equal in the energy they contained. The different effectiveness of the lights must, therefore, depend primarily upon the wave-lengths, and upon the chemical substances on which the lights acted. These substances are generally assumed to be of several kinds, and consequently it would be natural to expect that light should affect them differently. The degrees of effectiveness of the several lights for the eye and the skin were, however, remarkably uniform.

Sensitiveness to differences in wave-lengths, i. e. to color, was therefore present in the skin, as well as in the eyes of toads, though somewhat reduced in the former. Blue light was the maximum stimulus in the production of responses, the other lights forming a decreasing series, until in the red the effect was hardly more than that of darkness. The effect of each light was specific, and due, probably, to specific chemical changes caused by each set of wave-lengths. These specific chemical changes depended primarily upon the wave-lengths, and secondarily upon the absorption of light, the energy content of the several lights playing no part.

V. Summary.

1. Sensitiveness to differences in wave-lengths is present in the skin of toads, as well as in their eyes.
2. Blue light is the most effective stimulus in the production of responses, while green, yellow, and red form a decreasing series, corresponding only roughly to their relative positions in the spectrum.
3. Red light, when used singly, is not much more effective than darkness in the production of responses, and when paired with other lights, this slight effectiveness is even more decreased.
4. The sensitiveness of the skin to differences in wave-lengths is less than that of the eyes. When single lights were used, there were more negative responses when the skin only was exposed, than when the eyes, or the eyes and the skin together, were exposed. In balanced lights there were more movements toward the less refrangible light of a given pair when the skin only was exposed, than in the other two conditions of exposure, except in the pairs of red with green and with yellow.
5. The reactions when the whole animal was exposed to the light showed the influences of the slight differences in sensitiveness of the eyes and of the skin.

6. When tested with a narrow beam of blue light, the skin of eyeless toads was equally sensitive on all parts that were tested.

7. The distribution of the effectiveness of the several lights did not correspond very closely to their relative distribution in the spectrum.

8. The effects of each light were specific, and due probably to specific chemical changes produced by each. These effects were, primarily, a function of the wave-lengths, and secondarily, of the absorption of the light.

9. The intensity in the several lights used could have had no specific effect on the reactions of the toads to the different lights, for each light contained approximately the same amount or energy.

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Bulletin of the Museum of Comparative Zoölogy
AT HARVARD COLLEGE.
Vol. LIII. No. 6.

THE EFFECT OF COLORED LIGHT ON PIGMENT-MIGRATION IN THE EYE OF THE CRAYFISH.

By Edward C. Day.

With Five Plates.

CAMBRIDGE, MASS., U. S. A.:
PRINTED FOR THE MUSEUM.
December, 1911.

No. 6.— CONTRIBUTIONS FROM THE ZOÖLOGICAL LABORATORY OF THE MUSEUM OF COMPARATIVE ZOÖLOGY AT HARVARD COLLEGE, E. L. MARK, DIRECTOR. No. 227.

The effect of colored light on pigment-migration in the eye of the crayfish.

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I. Introduction.

Until recently no very critical investigations have been made upon the effects of colored light on pigment-migration in the eye. Throughout the work which has been done on this problem heretofore there runs an error which, although recognized by a few, has been either neglected or else unsuccessfully coped with. This error consists in ascribing the effect of colored light solely to its wave-length and of ignoring the factor of intensity.

In order to separate the effects of color from those of intensity in monochromatic light it was necessary to have recourse to some special device for measuring the intensities of lights of known but different wave-length. The instrument with which this was finally accomplished was the thermo-electric apparatus of Boys, known as a radiomicrometer, to be briefly described later. With the aid of this instrument a fact was discovered which prohibited the use of mono-chromatic light obtained through colored solutions. Since the infra-red waves, so potent in thermal energy, penetrated all solutions tested, it was impossible to get a measure of the intensity of the colors uncombined with that of the infra-red, and it was therefore necessary to resort to spectral light.

When the physical obstacle of intensity had been overcome, attention was turned to biological considerations. The subject of the migration of pigment in the retina was suggested to me for study, and since it was still an unsettled question whether the quality of light did in reality have any influence upon this phenomenon, the field seemed to be one full of promise. The problem was accordingly formulated as follows: — to determine whether different regions of the spectrum, when reduced to equal intensity, were equipollent in eliciting the migration of pigment; and if they were not, to seek, as a corollary to the problem, some quantitative expression for the difference in efficiency between the red and blue ends of the spectrum. The research was carried on in the Zoölogical Laboratory of Harvard University under the supervision of Prof. G. H. Parker, to whom I am indebted for much in the way of valuable suggestion, kindly criticism, and encouragement throughout the work.

II. Historical Review.

The vertebrates which have served in the study of pigment migration in the eye are fishes, doves, and especially frogs; while among the invertebrates the crayfishes, moths, and butterflies have been experimented upon. Between the years 1877, when Boll (**'77**) discovered the pigment-migration in the vertebrates and 1894, when Kiesel (**'94**) worked upon a moth, Plusia gamma, the frog chiefly has served for the study of the efficiency of colored lights in evoking this phenomenon. The only work ever done on the crayfish was that performed by Bell (:06) to ascertain how that animal reacted to colored lights and how changes in the retinal pigment affected the reactions of the animal to white light.

Contemporaneous with Boll's discovery that the pigment changed position under the influence of light, was his observation that the pigment-epithelium adhered to the retina least after exposure to red light, more after yellow and most after green, blue, violet and white light. Angelucci ('78) said, "Diejenigen Netzhäute welche anstatt in der Dunkelheit in einer möglichst intensiven rothen Beleuchtung verweilt hatten, verhalten sich in Bezug auf die Vertheilung der Pigmentkörner gerade so wie die Netzhäute der Dunkelfrösche." Blue, on the other hand, he reservedly stated, elicited greater migration than did white light. Engelmann ('84) claimed that the results of the work done by Van Genderen Stort, his student, and by himself with the use of light filtered through colored glass and by tests with spectral light, pointed to the probability that blue was the most effective. Three years later, however, Van Genderen Stort ('87) advocated green as being the most potent stimulus. That color, he observed, caused the pigment to proceed past the ellipsoids of the rods, the migration-terminus for ordinary light, out to the external limiting membrane.

In all of the previous investigations the factor of intensity had been neglected. It is obvious that if the purpose of experiment be to ascertain the effect which quality, *i. e.* wave-length, of light has upon the visual organ, then the quantity, *i. e.* amplitude of the light-wave or radiant energy, must be kept the same in each color used.

Pergens ('99) was the first to realize and give recognition to the claims of this argument. Discovering that blue of the spectrum was too feeble for measurement he resorted to combinations of colored glass with which to produce monochromatic light. The intensities of the colors were equated by means of a Ritchie photometer. Two years previous he had obtained results from experiments on Leuciscus, in which red stood last in rank of efficiency, green next, then yellow, and blue first. Continuation of his work upon this fish with supposedly measured light yielded different results. With a light-intensity of one "Hefnerkerze" he got a graded series of migration through red, yellow, green, and blue in ascending order. For lower intensities, however, the order became green, red, yellow, blue. The green, as indicated by his curves, evoked hardly any migration whatever. When the intensity was diminished until no migration occurred for any of the colors, Pergens found that the cones still responded, thus confirming Engelmann (**'84**) in the belief that pigment-migration and contraction of cones were independent of each other for low intensities of light.

The only other investigation of the photokinetic phenomena in the vertebrate eye under colored light is that of Herzog (**:05**). The aim of his research was not so much to ascertain the qualitative effects as the quantitative, viz., the effects evoked by different intensities of the same color. Colored solutions were used as light-filters. The intensity of any given color could be varied by changing the voltage of the white light by means of a rheostat; but the intensities of different colors were compared by judging the distinctness of an object as seen through the filters. The contraction of cones was the major, the migration of pigment the minor object of investigation. He reached the conclusion that the contraction of cones was a function of the intensity and also of the wave-length of light; and incidentally he found that blue was most and red least effective in eliciting the migration of pigment.

The literature on invertebrates begins with Kiesel's (**'94**) contribution on Plusia gamma. This investigator was the first in studying the influence of colored light upon the photomechanical changes in the visual organ to make use of the long-known phenomenon of glow in the dark-adapted eyes of arthropods. His observations on the effect of spectral light are summed up in one rather remarkable statement, "Nach meinen Zahlreichen, allerdings mit unzulänglichen Mitteln angestellten Versuchen scheint es, als ob ausser den uns sichtbaren Strahlen auch Ultraroth, das wir bekanntlich nicht sehen, eine Pigmentverschiebung bei Plusia gamma verursache, dass also auch ultrarothe Strahlen von diesem Thiere wahrgenommen werden." The observations of Bell (**:06**) on the crayfish have been mentioned. The most recent work on the problem is that of v. Frisch (**:08**) performed on moths and butterflies. Taking advantage of the phenomenon of glow, he made a series of tests with extirpated, dark-adapted eyes, placing them by couples in different regions of a band of spectral light and timing them for the extinction of the glow. This occurred first in the violet, next in the blue, but no difference could be detected for the rest of the spectrum in the time required for this change.

From the work of Boll in 1877 to that of Bell in 1906 these researches into the problem of photomechanical changes induced in the eye by colored light have been carried on (and here the work of Pergens, **'99**, can not be excepted) with the same, persistent error of method: the dilemma of quality or quantity, of color or intensity thus far had not been resolved.

In 1907 Hertel (:07) emphasized a fact of fundamental significance. He pointed out that photometric determinations of the strength of light are insufficient, because we do not know whether the visible energy measured by the photometer represents the total radiant energy which falls on the retina from a given illumination. A thermoelectric device was used to determine this. He remarked that, if one got by this method the amount of energy necessary for perception in our eye, then one was justified in putting the light-reactions of a subjective (light- or color-perception) and an objective (photomechanical processes in the retina) nature into closer relation, because both are functions of the total energy measured in the same units. After determining the amounts of thermal energy necessary to produce the sensations of red and blue as such (and they approximated each other closely, hovering around 8–10 degrees Celsius), the effect of the colors of the same intensity was studied in the matter of cone-contraction in the retina of the frog. Red produced less contraction than blue. In the similarity of the two energy-values for the perception of color and the contraction of cones, the author found support for the view that the latter played a rôle in the former. Hertel made no observations upon pigment changes for colors of equal intensity.

In the results obtained by Boll ('77), Angelucci ('78), Engelmann ('84), Van Genderen Stort ('87), Pergens ('99), Herzog (:05) upon vertebrates, and by Bell (:06) and v. Frisch (:08) upon invertebrates, one finds a general coincidence of opinion that red light is least effective on the eye; while Kiesel ('94), with his claim for infra-red as a stimulus, stands alone in the opposition. All except Van Genderen Stort regarded blue or violet as the most potent region of the spectrum. In direct conflict with Van Genderen Stort's observations of extreme migration under the influence of green, are the negative results of Pergens ('99) for the same color. The intermediate spectral regions, therefore, are of doubtful status. Notwithstanding the concord of opinion relative to the extremes of the spectrum, the subject was still open to question on account of the inaccuracy of method involved. The subsequent pages contain the results of an investigation of this problem after the intensity of the colors had been made constant by the aid of the radiomicrometer.

III. Experimental Investigations.

A. PRELIMINARY MATTERS.

1. APPARATUS. The apparatus, Fig. A, for furnishing the colored light consisted of a long rectangular box containing near its middle a 5 inch biconvex lens, *o*, with focal length of 28 inches; a Nernst light, *l*, situated in the rear at one focal point; and a prism-bottle, *p*, containing carbon bisulphide, located about six inches nearer the lens than the focal point at the front end of the box. In dimensions the box was six feet long, a foot wide and about a foot deep. In order to adjust the apparatus to the new direction taken by the light after its passage through the prism, the front end was given an angle of deviation from the main axis of about 45 degrees. At the point *d*, where the spectral band came to a focus, a small wooden case, Fig. B, grooved in floor and roof for receiving cardboard diaphragms, was situated. The grooves were so cut that they intersected the spectrum at the foci of the red, yellow, green, and blue-violet, respectively (*cf.* Fig. A). Thus when a diaphragm was inserted in its appropriate groove, a vertical slot in it coincided with the spectral region desired to be used, allowing passage to it while the rest of the spectrum was obstructed.

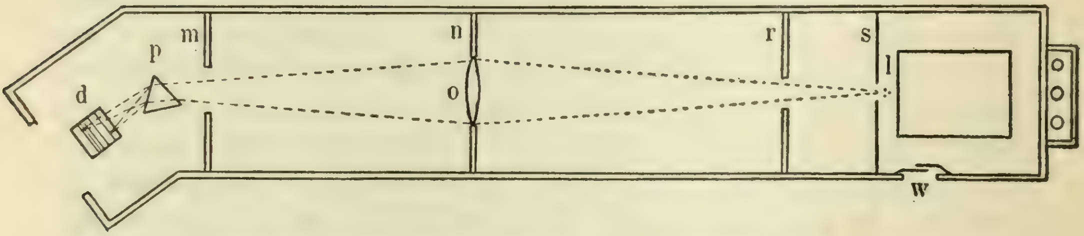

Fig. A. Plan of colored-light generator. *d*, diaphragm box; *l*, Nernst light; *m*, wooden diaphragm; *n*, diaphragm at lens; *o*, biconvex lens; *p*, prism; *r*, wooden diaphragm; *s*, iron diaphragm; *w*, window.

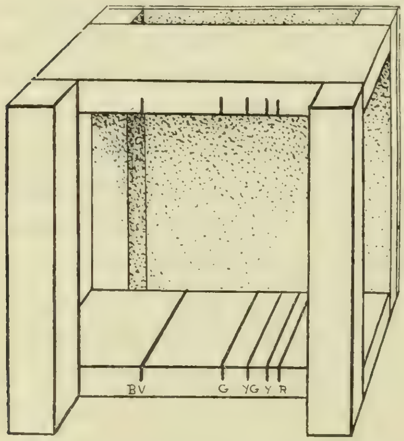

Fig. B. Diaphragm box showing the positions at which the diaphragms for blue-violet (BV.), green (G.), yellow-green (YG.), yellow (Y.), and red (R.) were slipped in.

By regulating the width of the slot in each diaphragm the radiant energy in the various regions of the spectrum could be reduced (*e. g.* in red) or enhanced (*e. g.* in blue-violet) until an equality, as determined by the radiomicrometer, was established between them. Besides the coat of dead black paint on the interior of the box, three diaphragms, two permanent ones of wood at *m* and *r*, Fig. A, and a removable one of sheet-iron at *s*, reduced internal reflections from the walls to a minimum. A small window, *w*, on one side and near the rear, together with an aperture cut in the lid and surmounted by a metal chimney, served as ventilators to reduce the heat from the lamp. Metal flanges in the chimney and at the side aperture were so overlapped as to prevent any appreciable leakage of light. By means of a metal peg projecting upward from the table and inserted in a hole in the floor of the box at a point directly beneath the center of the prism, the apparatus was so pivoted that the beam of monochromatic light could be directed at will.

Illumination was furnished by three 220-volt Nernst filaments of the pattern used in the Schwann lamp and so arranged that they might be used either in combination of three, for blue-violet and green, two, for yellow, or singly, for red light. The feebleness in radiant energy of the blue end of the spectrum necessitated not only a wide slot in the blue diaphragm but, in addition, this reënforcement by triple combination of glowers to obtain sufficient energy to equal that furnished by the single glower and a narrow diaphragm-slot in the red. A front perspective view of the lamp devised for facilitating these combinations is shown in Fig. C. At one end of a wooden base, *a*, was erected an arch of brass, *b*, about six inches high. The three L-shaped posts, p, p', p'', and the arms, r, r', r'', above were also of brass. The middle arm, *r*, and post, *p*, were screwed firmly to the underside of the brass arch and to the wooden base, respectively. On the right side of this central combination, *rfp*, the arm r' was fastened to the arch with a thumb-screw and nut so placed that the pivoting point *m* plumbed exactly with the screw *n* in the post p' below. The wooden column *h*, sheathed with blackened asbestos, connecting the backward extensions of r' and p', so firmly united these two parts that they could be rotated coördinately about *mn* as an axis. Whereas the upper platinum connection of the glower f' was wedged into a hole in the arm r' with a copper plug, *e*, the lower connection, in order to allow for the expansion of the filament when heated, was suspended freely in a mercury-filled cup, *c*. This brass cup was bound with copper wire to the post p'. The whole system

$r'f'p'h$ was duplicated on the opposite side in $r''f''p''$. When the lateral glowers f' and f'' were swung into the center, the three formed a compact prismatic combination, which generated the powerful illumination necessary for the blue-violet and green. A cross-section of this combination is represented in the plan of the apparatus, Fig. A, by the triangle of three dots at l. For yellow only f and f' were needed, while the single, central glower was sufficient for red. In the latter two cases the unused glowers were swung out of the field to avoid the disturbing reflections from them.

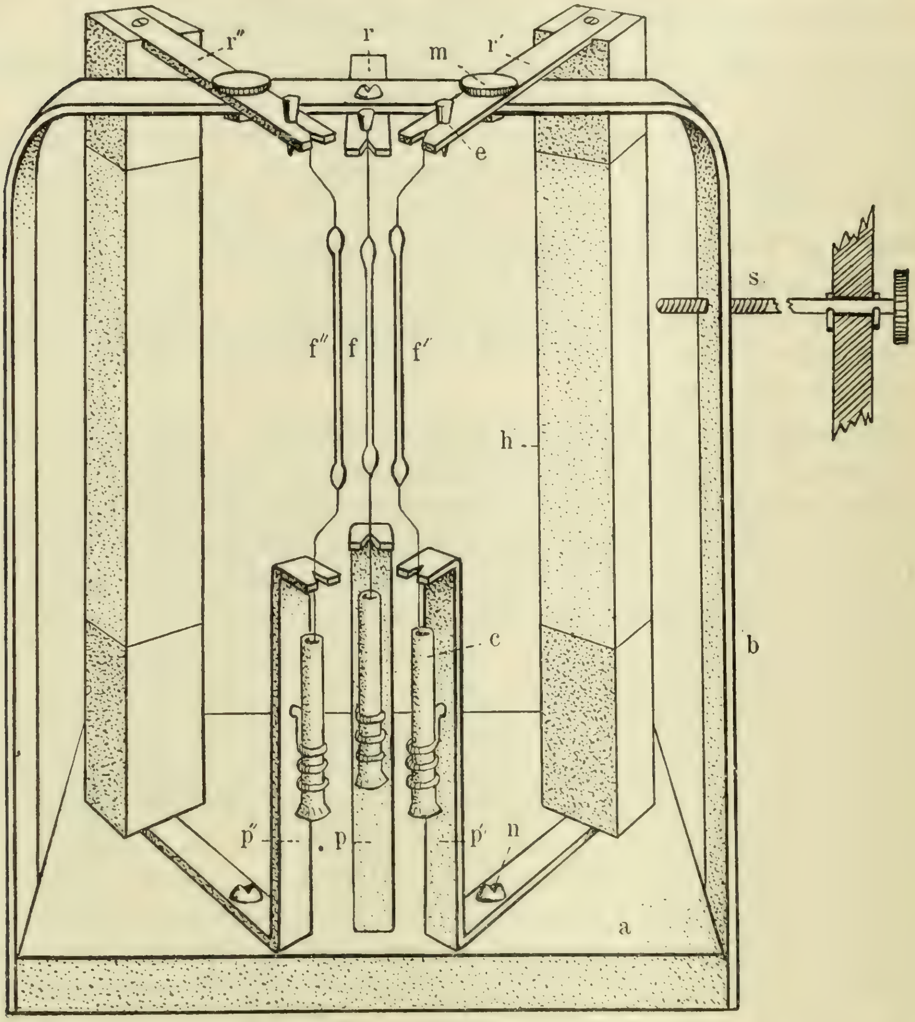

Fig. C. Front perspective view of lamp. *a*, wooden base; *b*, arch of brass; *c*, mercury cup; *e*, copper plug; *f*, *f'*, *f''*, Nernst glowers; *h*, wooden column; *m*, pivoting point; *n*, screw; *p*, *p'*, *p''*, posts; *r*, *r'*, *r''*, arms; *s*, adjusting screw.

The wiring of the lamp is represented diagrammatically in Fig. D. One branch, *r*, of the circuit ran to a leg of the brass arch of the lamp thereby supplying the upper ends of the glowers, while the other branch, *s*, went via the switches *a*, *b*, *c* and the ballasts *d*, *e*, *f*, to the back extensions of the three brass posts which connected with the lower ends of the glowers. Switch *a* controlled the whole circuit. The accessory switches *b* and *c* were introduced so that either or both of the lateral glowers might be extinguished without interrupting the circuit for the remaining ones when a different region of the spectrum was to be used.

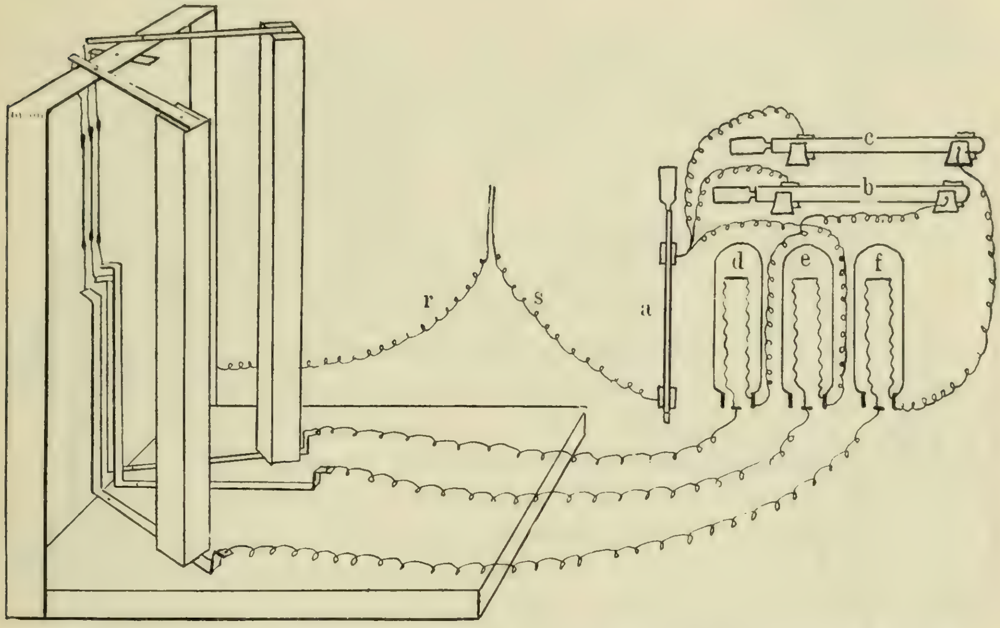

Fig. D. Diagram of wiring of lamp. *a*, *b*, *c*, switches; *d*, *e*, *f*, ballasts; *r*, wire to brass arch; *s*, wire to the ballasts.

At this juncture it may be appropriate to remark briefly upon the method by which equal intensity of the colors was obtained.

The radiomicrometer, reduced to simplest terms, is a slender loop of wire containing one or more thermal junctions, suspended by a delicate quartz filament to hang within a magnetic field. In Fig. E the loop, *l*, was of fine copper wire; the thermal junction was a small, blackened platinum disk, *p*, soldered into the loop with a fusion of bismuth and antimony, *s*; the suspension, *f*, was an extremely fine quartz filament; and the magnetic field was furnished by a horseshoe magnet, *h*. The principle by which the radiomicrometer operates is in general this:— if a beam of light falls upon the thermal junction (via aperture *a* in side of box), an electric current is generated in the circuit; and since a wire carrying a current is surrounded by a magnetic field, this loop, being converted into a temporary magnet and free to rotate will adjust itself like a magnetic needle to the influence of the stationary magnet. The amount of rotation is proportional to the radiant energy of the light which strikes the thermal junction, and can be measured on a scale reflected into a telescope by a tiny mirror, *m,* included in the suspended system. By this instrument the intensity of the colored lights was registered in the same units. After reducing a color to the common thermal equivalent, it was examined with a spectroscope and its spectral range was recorded upon the diaphragm used. Whenever a Nernst filament was replaced and the lights were again balanced on the radiomicrometer, these spectral ranges had to be modified slightly. Through the first part of the investigations four spectral regions were employed whose ranges in wave-length were:—

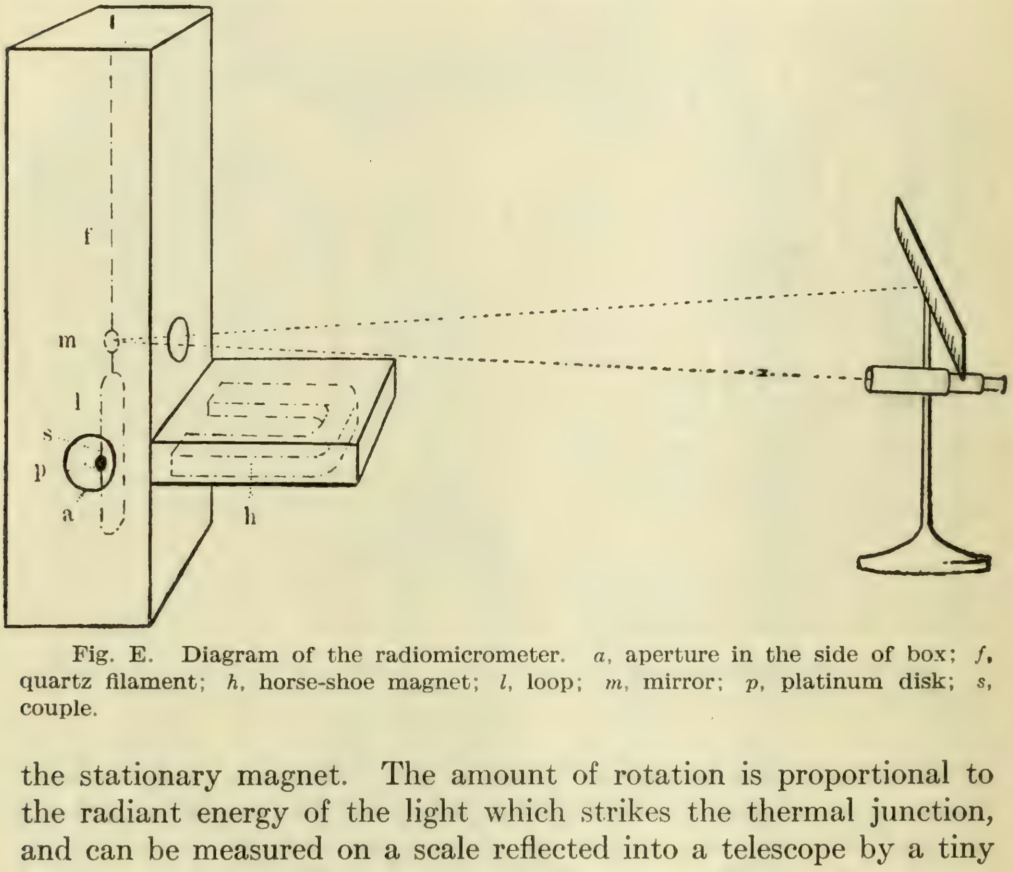

Fig. E. Diagram of the radiomicrometer. *a*, aperture in the side of box; *f*, quartz filament; *h*, horse-shoe magnet; *l*, loop; *m*, mirror; *p*, platinum disk; *s*, couple.

Blue-violet	430–490 $\mu\mu$
Green	495–545 “
Yellow	565–620 “
Red	625–665 “

but for the later experiments three regions, which centered upon 460, 550, and 645 approximately, making intervals of 90 $\mu\mu$ between them, were used:—

Blue-violet	430–490 $\mu\mu$
Yellow-green	524–576 "
Red	625-665 "

If all factors concerned, the lamp, lens, prism, and diaphragms had retained a constant position in the apparatus, then the different regions of the spectrum would always have coincided with the slots in their appropriate diaphragms; but, owing to slight shifting of the glowers due to expansion and contraction, it was found necessary to introduce an adjusting screw, *s*, Fig. C, by the manipulation of which from outside the box, the whole lamp could be moved laterally, *i. e.* in a direction at right angles to the long axis of the light-box, until the exact spectral region, as determined by the spectroscope, was obtained through the slot in the diaphragm. Prior to the employment of each color the light was thus set by means of the adjusting screw and spectroscope.

The only other pieces of apparatus required were vessels in which the animals could be kept in absolute darkness for several hours, and a few diaphragms which could be interposed at intervals between the animal and the front of the light-box. Tin troughs about three and a half feet long, eight inches wide and four inches deep, when coated with black wax on the inside and provided with light-tight covers served the first purpose; while screens of opaque cardboard served to eliminate any extraneous reflected light during the exposure of the animal to the monochromatic light.

2. ANATOMICAL ORIENTATION. The animal used in all of my experiments was a common species of crayfish, Cambarus affinis. There were several advantages which, in comparison with the vertebrates commonly used, made this animal a favorable subject for experimentation; although not indigenous in the neighborhood of Cambridge, it easily adapted itself to the aquarium after importation from the South; its size was convenient for manipulation; the migration of pigment was pronounced, and especially was it superior for the ease with which the eyes could be instantly fixed after exposing them to the colored light.

A brief survey of the topography of the compound eye will be serviceable with respect to the problem in hand. For a detailed account of the anatomy one may turn to the description of the European species, Astacus, by Parker ('**95**). The structure of the eye of

Cambarus is essentially the same. Exteriorly the eye has the form of a stalk encased with tough cuticula and capped by a dome whose outer surface is the thin transparent continuation of the cuticula of the stalk. On the interior of the stalk, proceeding distad from its base, are the optic nerve, four large ganglia, and the retinal fibers which ascend from the ganglia to the retinal cells. Partitioning the stalk off from the dome is the basement-membrane or membrana fenestrata. Between the periphery of the dome and the basement-membrane lies the dioptric and receptive apparatus made up of many radial units or ommatidia. The components of a single ommatidium (*cf.* Plate **1**, Fig. 1b) in centripetal order are these: — a corneal facet in the outer cuticula (*ct*); two subjacent corneal hypodermal cells (*cr*); the cone (*c*) of four cells, differentiated into a distal, highly refractive part and a less refractive proximal portion; the spindle-like rhabdome (*m*), which abuts on the basement-membrane (*bm*); then, flanking the rhabdome are the seven proximal retinular cells (*pp*), while encompassing the outer segment of the cone are two distal retinular cells (*dp*), both sets of which, proximal and distal, contain brownish-black pigment-granules; finally, two sets of yellowish accessory or tapetal cells, a distal one lying between the distal retinular cells and the cone, and a proximal set (*t*) situated, with respect to the rhabdome, outside of the proximal retinular cells. Of these ommatidial components the cone is regarded by Parker (**'95**) and Hesse (**:01**) as the dioptric apparatus, while the rhabdome is considered to be composed of differentiations of the proximal retinular cells and to be the receptive organ. It is only through this latter interpretation of the function of the rhabdome that the phenomenon of pigment-migration finds its significance. In Cambarus the rôle which the distal retinular cells play in the photomechanical changes of the eye is slight as compared with that of the proximal retinular cells. All my experiments dealt, therefore, with the influence of colored light upon the migration of pigment in these proximal cells alone. I shall often make use of the terms "retinal cells" and "retinal pigment" in referring to this set of cells.

In order to establish a point of departure it was necessary to acquaint myself with the typical condition exhibited by an eye subjected to dark and by one subjected to light. Two standard eyes were prepared, one from an animal kept in absolute darkness for six hours and killed in the dark, the other from an animal exposed for six hours to bright, diffuse daylight and killed in the light. Microscopic examination of the two showed diverse conditions. In the dark-eye (Plate **1**, Fig. 2b) the retinal pigment lay almost entirely proximal to the basement-membrane (*bm*) packed closely against it and extending inwards from it in a dense mass. Blunt processes of the pigment (*rp*) could usually be seen projecting into the tapetal layer (*t*) distal to the basement membrane; in fact I never obtained a condition such as is shown by Parker's (**'95**) preparations of Astacus, where no retinal pigment was to be found in the dark-eye distal to the membrane. In the eye exposed to light (Plate **1**, Fig. 1b) the pigment had moved out through the fenestrated membrane, to surround the rhabdomes — even creeping out laterally into the interstices between the rhabdomeric plates — and to become densely accumulated in the distal ends of the retinal cells. Proximal to the basement-membrane only a comparatively slight amount of pigment was left. In the dark, then, the pigment lies proximal to the fenestrated membrane while in the light it lies practically all distal to this.

3. Technique. For the most part the technique required for study of these photokinetic changes was simple. Since no killing nor staining fluids were needed the procedure for microscopic study was thereby considerably abridged. On the other hand the difficulty involved in removing the tough cuticula without seriously impairing the eye retarded the process. The method employed by Parker (**'97**) and Congdon (**:07**) was adopted for killing; by dropping the animal into hot water at 80°–85° C. the position of the pigment was instantly fixed by coagulation of the protoplasm. After the eye-stalk had been in 70 % alcohol for some time the cuticula was removed. This process was performed in a shallow dish of alcohol and best under a binocular dissecting microscope. By making an incision at the base of the dome with a sharp scalpel (with the knife-edge turned distally), a flap of the corneal cuticula could be turned up and, with a pair of fine forceps, be peeled off over the dome. After the dome had thus, bit by bit, been entirely peeled, it only remained to remove the tough cuticular casing of the stalk. The point of a fine needle was inserted at the base of the dome and worked carefully around the inside of the rim of the stalk-cuticula at the point where it had been girdled, to loosen the stalk from its attachment to the cuticula in that region. The cuticula of the stalk was then cut lengthwise along two sides thus releasing the eye *in toto* from its encasement. Embedding was done in paraffin, and the eye cut into sections having an antero-posterior direction and a thickness of 10 micra. A few sections from the middle of the eye were mounted, unstained, in balsam for study. The amount of migration was judged by the distance traversed between the basement-membrane and the nuclei (visible although unstained) in the outer ends of the retinal cells.

B. Observations.

1. Section method. A series of preliminary tests was made to ascertain the time required for the pigment to attain the complete recessive condition for dark-adaptation. The probability is that it varies with the physiological state of the animal, being more rapid for a vigorous individual than for one in poor condition. Although four hours were found to suffice, yet, for the sake of certainty, the period allotted for dark-adaptation was six hours.

In the experiments with colored lights two methods were employed:— the first consisted of exposing the crayfish to the color, killing it instantly in hot water and preparing the eye for microscopic study; while by the second a comparison of the influence of the different colored lights in extinguishing the glow of the living eye was made by direct observation.

(a). *Procedure.* In employing the first method the animals were exposed at different distances from the source of light and for different periods of time. The crayfish was submerged in a tank of water just far enough to permit respiration but not to cover the eye during the exposure (Fig. F.). At first the animals were held by a clamping device, but later by hand. When a strong intensity and long exposure (one hour) were tried, sections of the eyes revealed no detectable difference in the influence of the several colors because the amount of migration was the same in every case. The intensity was then diminished by moving farther away from the light, and simultaneously the length of time for the exposure was also reduced. At $\frac{1}{25}$ the original intensity and for an exposure of fifteen minutes the efficiency of the red showed slight signs of weakening; and at $\frac{1}{668}$ of the initial intensity, *i. e.* at 550 cm. distance, or as far away from the light as the crayfish could be placed, the red was decidedly behind the other colors in effect, while the blue-violet, green and yellow remained about equal. To differentiate the latter was the next problem. The exposure was cut down to one minute. A stop-watch together with precautions for eliminating delay between the end of the exposure and the immediate fixation of the pigment *in situ,* refined the process to the desired degree. Although the three colors could not all be separated, nevertheless the efficiency of yellow and green, as yet on a par with each other, could be differentiated from that of the more potent blue-violet.

(b). *Results.* The results obtained by the section-method are given in Table I. The nine series, each including usually four animals, were run upon separate days. In each series a single animal was exposed to a single color, and a check animal which had been confined in the same dark chamber was killed in the dark at the end of the experiment. On account of the irregularities in the

TABLE 1.

Amount of migration shown by sections.

Values under BV, G, YG, Y and R (blue-violet, green, yellow-green, yellow and red, respectively) are estimates of the distance traversed between the basement-membrane and the row of nuclei of the retinal cells expressed in tenths of that distance. The last column gives similar values for animals kept in the same dark chamber and killed in the dark after the others of the series had been exposed. In the summary, read horizontally, each color is compared with BV and G with Y, showing the number of times one was ahead of the other in efficiency.

Series	BV	G	YG	Y	R	Distance from Light box	Period of Exposure	Check
						cm.	min.	
1. ♂♂	7	8		7		550	1	5
2. ♂♂	8	6		8		550	1	5
3. ♂♂	10	8		7	6	550	1	8
4. ♀♀	10	8		9	7	550	1	2
5. ♂♂	8	8		9	2	550	1	4
6. ♀♀	8	6		3	4	550	1	1
7. ♀♀	7		8		9	550	1	3
					1	300	1	
8. ♀♀	6					550	1	2
					8	550	20	
					3	550	20	
					6	250	1	
					8	250	1	
9. ♀♀	7		5			550	1	
	6		2			550	1	

Summary	Number of times ahead					Tie	Total Trials
	BV	G	YG	Y	R		
BV *vs.* G	4	1				1	6
" " YG	3		1			2	6
" " Y	2			1			3
" " R	9				1		10
G " Y		3		3			6

boundary of the advancing pigment, a general visual comparison of the slides under the microscope afforded a better basis for judgment than could be obtained by actual measurements. The values given in the columns under BV, G, YG, Y and R (standing for the colors blue violet, green, yellow-green, yellow, and red, respectively) were estimates, on a scale of ten, of the distance the pigment had migrated, regarding the basement-membrane as a starting point and the row of nuclei in the distal ends of the retinular cells as the terminus. Thus, in series 3 the blue-violet occasioned the pigment to migrate the whole distance and was evaluated at 10, while red elicited it for only 6 tenths the distance. This series, which grades with respect to influence on migration from blue-violet through green and yellow to red, is photographically reproduced in Plates **2** and **3**. In Fig. 3, Plate **2**, which exhibits the effect of blue-violet, the pigment is out around the nuclei of the retinal cells; in Fig. 4, Plate **2**, (effect of green) it has migrated slightly beyond the boundary (the broken line in left half of photograph) of the tapetal layer, but not so far as the row of nuclei (indicated by dotted line); in Fig. 5, Plate **2**, (yellow) the pigment and tapetum are coextensive; but in Fig. 6, Plate **3**, (red) the pigment, though coextensive with the tapetum at the center of the retina, falls short of the outer boundary of the tapetum on the sides (as the left half of the photograph has been retouched to indicate). All retouching for sake of emphasis, has been restricted to the left half of each photograph.

The summary of results in the lower portion of Table I shows, reading horizontally, that for BV vs. G, out of 6 trials (a trial being a comparison of two animals in the same series), blue-violet showed the greatest migration 4 times; for BV vs. YG, blue-violet was more effective 3 times, yellow-green once, while twice they tied; for BV vs. Y, blue-violet was more effective 2 out of 3 trials; but for G vs. Y, each evoked more migration that the other 3 times out of 6. In the case of BV vs. R, allowing for the fact that in series 7 and 8 certain modifications were made in exposing to red, increasing either the intensity or the time of exposure in order to obtain a migration equivalent to that with blue-violet, it is maintained that, including these with the other series, the blue-violet outweighed the red in efficiency 9 out of 10 times. In addition to this there were six preliminary series in which red failed to evoke as much migration of the pigment as the other colors did.

The evidence furnished by these observations is conclusive only for the difference in efficiency between the extremes of the spectrum.

As regards blue-violet, green, and yellow, what evidence there is points to the fact that they stand close together, but of the three blue-violet is possibly the more efficient in eliciting the migration of pigment.

Having discovered a decided difference in the efficiency of blue-violet and red, it was suggested that some quantitative expression be sought for that difference: —how much more potent was the blue-violet than the red? The attempts to obtain an answer by the section-method are shown in the variations of the exposures to red in series 7 and 8; but they are too few to be even indicative. This question was investigated and carried through to a conclusion by the second method employed in the experiments, viz. by direct observation of the changes in the glow induced by red and by blue-violet light, respectively, in the dark-adapted eye.

2. DIRECT-OBSERVATION METHOD. (a) *Phenomenon of glow.* The fact that the phenomenon of glow had been observed by Lowne (**'84**) in moths and butterflies, by Exner (**'91**) in various insects and Crustacea, and by Kiesel (**'94**) in a moth, led me to investigate the matter in the crayfish. In this animal, too, it was found that the dark-adapted eye was quite different in aspect from the one adapted to light. In the daytime the eye (Plate **1**, Fig. 1a) presented a dark central spot framed by a lighter peripheral area. As early as 1855 this phenomenon had been described in Limulus by Leydig and the term "Pseudopupille" applied to it from its apparent analogy to the pupil in the vertebrate eye. Exner (**'91**) has investigated the phenomenon among many arthropods and has offered an explanation for it involving the principle of the cylinder lens, realized in the cones, and also the rôle played by the pigment and tapetal cells.

At night the pseudopupilla in the crayfish had vanished and the eye, when examined by a flash-light, appeared no longer dark, but glowed with a bright, metallic-orange light (Plate **1**, Fig. 2a); and when illuminated in the aquarium, the eyes of the crayfish shone out of the darkness like beads of fire. It was not a case of fluorescence but of reflection. Since in the dark-eye the pigment of the retinal cells occupied a position proximal to the basement-membrane (Plate **1**, Fig. 2b), the retinal tapetum was left exposed, and it acted as a reflector for all light entering the eye. The rudy orange hue was due to the presence of a red substance in the rhabdomes,—observed by Lowne (**'84**) in the cabbage-butterfly, by Parker (**'95**) in Astacus and by myself in fresh maceration preparations of Cambarus—, possibly a "visual-red"; and since the rays had to pass through this before they struck the reflecting layer, they were returned as filtered light. After exposure to light for several minutes the original metallic-orange faded to a dull yellow hue and, *pari passu* with this change, there appeared a dark area in the center (*cf.* Plates **4**, **5**, Figs. 7a, 8a, 9a). The alteration of color is believed to be due to a partial bleaching of the visual-red substance in the rhabdomes. The darkening of the center of the eye is explained as the result of the migration of the pigment to cover the retinal tapetum (Plate **5**, Fig. 9b), whereby reflection is prevented. Through acquaintance with this fact, it was conceived that a judgment might be made as to the amount of darkening corresponding to the difference in influence of the colored lights.

(b). *Procedure.* Individual records for each crayfish were kept and contained the following: — the sex; exposure data including color of the light, intensity in terms of centimeters distant; the appearance of the glow at the beginning and at the end of exposure; period of exposure; and temperature of the water. Since the initial glow could be seen in the colored light, every animal was rejected from a given experiment if the eye was not in full glow. The animals were put in the dark at noon and left until evening, when the experiments were conducted. The reason for this procedure was to obtain the glowing condition as nearly as possible in accordance with the normal change induced in the eye by the transition from day to night; in other words, if there were any physiological rhythm in the pigment-migration, such as Kiesel ('**94**) observed in Plusia gamma, my object was to work with, and not counter to it. Whether the migration is periodic in the crayfish I can not say. So far as the evening hours were concerned, the eyes of the animals were then nearly always in good glowing condition.

The arrangement for making the exposure is shown in Fig. F. The crayfish was held by hand partly submerged in a pan of water in such

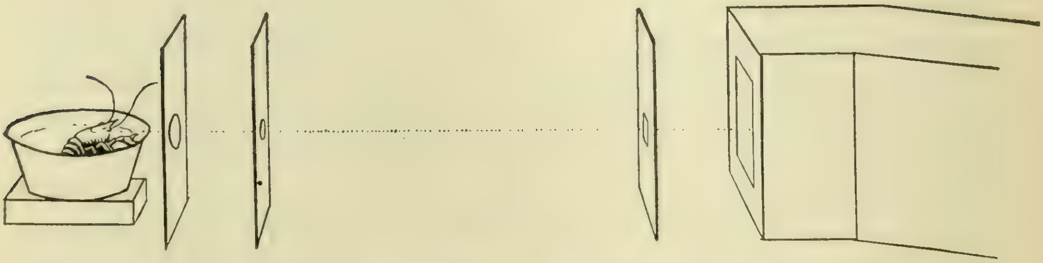

Fig. F. Diagram of apparatus for exposing crayfish to colored light. At extreme right is the end of the colored-light generator; at the extreme left a dish containing the crayfish; between these positions are three diaphragms.

position as to expose one eye only to the direct action of the light, and with two or three diaphragms interposed between it and the source of light to exclude all reflected rays. At the close of the exposure, which was timed by a stop-watch, the eye was examined by focusing a flash-light on it, a judgment was made by direct observation of the amount of darkening in its center, and the condition was sketched into a standard blank circle representing the eye, stamped in the record book. The coloration of the eye was also indicated in the circle by means of colored pencils. The eye on the side of the head away from the light, since it retained its glow and showed no dark center (when the exposure did not exceed five minutes), served as a check upon the other and was recorded in like manner.

(c) *Preliminary tests.* The first step, as in the section method, was to make preliminary tests for differences in the effects of the three chosen spectral regions, blue-violet (430–490 $\mu\mu$), yellow-green (524–576 $\mu\mu$), and red (625–665 $\mu\mu$). As before, many trials were necessary to find the most favorable combination of intensity and exposure to elicit perceptible differences in the external appearance of the eye correlated with the migration of pigment. Whereas in the spring, when the first method had been applied, a weak intensity and short exposure had yielded the desired results, in the winter, when the second method was employed, the animals, being in a more sluggish condition, required a much higher intensity and longer exposure. The outcome of the preliminary observations confirmed those obtained from the sections: — the blue-violet was decidedly more potent then the red, but the case of blue-violet *vs.* yellow-green was doubtful, since they were so close in efficiency.

(d) *Effects of blue-violet and red compared.* The method was then concentrated upon the question of obtaining a quantitative expression for the greater efficiency of the blue-violet as compared with red: first, by varying the intensity while keeping the time of exposure constant, and secondly, by varying the time while keeping the intensity constant.

(1) Intensity varied. Ten minute exposures were made to blue-violet at a distance of 200, 250, 300, and 400 cm., and to red at 50, 100, 150, and 200 cm., with the expectation that positions giving equivalent migration in the blue-violet and the red might be obtained. These distances were measured from a point situated several centimeters in front of the light-box and corresponding to the position of the radiomicrometer when the lights had been balanced in intensity. It was found that the full number of exposures could not be made on

successive days on account of deterioration manifested in the eye by a gradual diminution in the migration, so that usually only one, or sometimes two, legitimate matchings in amounts of migration evoked by the two colors could be obtained for each animal.

In order that the results might be compared and summarized in some way, it was necessary to assign a quantitative value to the amount of migration exhibited by each recorded sketch after comparing it with an arbitrary standard. This standard was a circle, made with the same stamp as those in the records, with a dark area in the center representing a medium amount of migration which, based on a scale of 5, was evaluated at 3. Each migration-record, therefore, was referred to this standard, judged for its relative amount of migration, and assigned a value accordingly. Table II contains these evaluations in the vertical columns under the distances at which the exposures were made (50, 100, etc. cm.). In the right hand portion of the table are given those distances — deduced from the assigned values either directly, where one in red équalled one in blue-violet, or indirectly by interpolation — at which red is probably equivalent to blue-violet in eliciting the migration. Discretion had to be exercised in determining the validity of evaluations chosen for comparison, because, owing to deterioration in the response of the pigment according to the sequence of the exposures, certain cases apparently equal in the table were quite incomparable. Taking animals K and L for example, the exposures for both were in the following sequence:— to BV at 400 cm. (Jan. 24), 300 cm. (Jan. 26), 200 cm. (Jan. 27); to R at 150 cm. (Jan. 30), 100 cm. (Jan. 31), 50 cm. (Feb. 1), the progress through each color being from the lower to the higher intensity. Although two days of rest were allowed between the last exposure to blue-violet and the first to red, yet the exposure on the third consecutive day to red showed evidence, if one compares the values for 50 cm. with those at 100 cm., of exhaustion in the eye. Again, in animal 38, where red was tested at 50 cm. at the beginning and again at the end of the series, this deterioration was manifested. Final deductions from the assigned evaluations were made, therefore, after the sequence of exposures had been taken into consideration. The distances at which the efficiencies of the two colors were on a par were averaged and equated, thus R at 107.5 cm. = BV at 247 cm. Since the latter distance was 2.29 times the former, the intensity of the former was $(2.29)^2$ times the latter, *i. e.* BV = 5.2 R in terms of power to elicit the migration of pigment.

In order to verify the results obtained by this necessarily artificial

TABLE II.

Intensity (i. e. distance) varied.

Time of exposure constant (10 minutes). Evaluations were made with reference to a single arbitrary standard of medium migration. Last two columns give distances corresponding to equivalent migrations for red and blue-violet as deduced from the assigned evaluations. (Evaluations in parentheses were ignored in making the deductions, because they represented migrations after the eyes had deteriorated in value.) From averages of these distances was computed the relative intensity of BV in terms of R, given in Equation I. Equation II was obtained after rejecting dubious cases.

	Red				Blue-Violet					Distances for Equivalent Migration	
Distance in cms.	50	100	150	200	150	200	250	300	400	R.	BV.
Animal										cm.	cm.
C ♀	3	2.5	2			3		1	0	150	250
										50	200
G ♀		2				2		0		100	200
K ♀	(1)	2.5	2			2		1	0	150	200
L ♀	(3)	4	3			4		3	1	100	200
										150	300
19 ♀		4				4				100	200
20 ♀			2	0		4	3			150	300
22 ♀	4	3	2			5	4			50	250
24 ♀			2	2			4	4		100	300
25 ♀	2	0				1	2			50	250
										100	200
28 ♀	5	4	2			5		4	2	50	200
										100	300
										150	400
35	3	2	0			1		1	0	150	300
										100	150
36 ♂	4	3	1						1	150	400
38 ♀	4(3)	2	1		3	1		0		50	150
										150	200
									Av.	107.5	247

$$\frac{BV}{R} = \frac{247}{107.5} = 2.29 \text{ in terms of distance.}$$

$$BV = (2.29)^2 \text{ R in terms of intensity, or}$$

Equation I: $BV = 5.2\ R$

Equation II: $BV = 5.6\ R$ after eliminating G, K, 24, 25, 35 and 36, whose records were less easily compared.

and devious method of comparison, a new and independent set of evaluations was made, given in Table III, for the same migration-records *i. e.* for those used in Table II. This time the standard of reference consisted of a graded series of migrations exhibited by animal 28 in three separate exposures to red at a distance of 150, 100 and 50 cm. This series was evaluated at 1.5, 3, and 4, respectively, and was used as a scale by which to judge the other records. Not only was the value of sequence weighed again, but the records of animals of dubious physiological status were marked and, in the averaging, were taken into consideration. When all the animals were averaged, the final equation was BV = 6.5 R; after rejecting animals G, K, 24, 25, 35, and 36, this became lowered to BV = 5.6 R. Returning to Table II, the elimination of the same animals raised the final result from 5.2 to 5.6, showing perhaps that these animals were so dubious as to make the results oscillate up and down. From Tables II and III the final expression, therefore, for the elicitibility of pigment-migration, determined by varying the intensity, was reduced to BV = 5.6 R approximately.

(2) Time of exposure varied. In this modification of the experiment, instead of making several exposure tests with both colors, only one exposure was made with blue-violet (for a five minute period), while three were made with red viz. for 20-, 30-, and 45-minute periods, with the expectation of securing amounts of migration less than, equal to, and greater than that evoked by the blue-violet.

Throughout this series of experiments the crayfish were usually exposed only every other day, in order to minimize the disturbing factor of deterioration. Table IV was compiled in a manner similar to that for Table II by evaluating with reference to a single arbitrary standard condition, while Table V was based, similarly to Table III, upon an independent set of values assigned relative to three graded and evaluated migrations from the records of animal 22. The average of each column of values under the respective exposure-periods was taken. Since the average evaluation for red at 30 minutes was smaller, while at 45 minutes it was greater, than that for blue-violet at 5 minutes, by interpolation the period of time at which red would have equalled the effect of blue-violet at 5 minutes was computed to be 36.8 minutes in Table IV, and 39 minutes in Table V, or, if averaged and reduced to the previous equation, BV = 7.6 R. Another determination, made from Table V by interpolating for the requisite periods of time in each individual set of

TABLE III.

Intensity varied. Time constant (10 minutes). Evaluations made from the same observation-records as in Table II, but with reference to three graded migrations exhibited by animal 28.

	Red.			Blue-Violet.					Distances Equivalent Migration.	
Distance in cms.	50	100	150	150	200	250	300	400	R.	BV.
Animal									cm.	cm.
C	2	1	1		1.5		0.3	0	75	200
									150	300
G	0.5				1		0		50	200
K	(0.3)	1	1		0.5		0.2	0	100	200
L	(2.5)	3	2.5		2		2.5	0.5	150	300
19		3.5				3.5			100	250
20			1		2.5				100	250
22	2.5	2	1.5			2			100	250
24			1			2.5			75	250
25	0.5	0	0			1.5			50	300
28	4	3	1.5		3.5		3	2	75	200
									100	300
35	1.5	1	0		0.5		0.1	0	100	200
36	2.5	2	0.5					0	150	300
38	1.5	1	0.5	2	0		0	0	50	150
								Av.	95	243.3

$\frac{BV}{R} = \frac{243.3}{95} = 2.56$ in terms of distance.

$BV = (2.56)^2$ R. in terms of intensity, or

Equation I: $BV = 6.5$ R

" II: $BV = 5.6$ R on rejecting G, K, 24, 25, 35 and 36.

records first and then averaging these, yielded BV = 7.2 R; and when this was included with the other determinations, the final average expression for blue-violet in terms of the efficiency of red, ascertained by increasing the period of exposure, became 7.4, a value which is somewhat higher than that previously determined (5.6) by varying the intensity.

TABLES IV AND V.

Time of exposure varied. Distance constant at 150 cm. Evaluations made with reference —

Table IV — to a single standard condition of medium migration,—
Table V — to three graded migrations exhibited by animal 22.

	IV.					V.					
Color.	Red.				BV.	Red.				YG.	BV.
Distance in cms.	150			50	150	150			50	150	150
Exposure in minutes.	20	30	45	5	5	20	30	45	5	5	5
Animal											
39. ♀			5		3						
39. ♀	2	1.5	4	5	2	2	2	3			2.5
40. ♂	1	3	3.5	3.5	3	1	2.5	3.2	2.5	2	2.5
41. ♂	3	3	5	5	3.5	2	2.5	4	5	2	3
42. ♂	1	2	2.5	4	3	0.5	1	2.5	3.5	2.2	2.5
43. ♀	1	3	3.5	3.5	3	0.5	2	2.5	2.5	2	2.2
44. ♀	1.5	2.5	3	4	3.5	1	2.2	2	3.5	1.5	2.5
48. ♂	1	3	3.5	5	3	0.3	2.5	3.3	4	3.5	2.5
52. ♂	1	2.5	3		3	1	2.5	2.7			2.5
22. ♀	1.5	2.5	3		3	1	2.2	3			3
Total	13.0	23.0	36.0	30.0	30.0	9.3	19.4	26.2	21.0	13.2	23.2
Average	1.4	2.5	3.6	4.3	3	1	2.15	2.9	3.5	2.2	2.6

A computation from the average evaluations for R at 30 minutes and 45 minutes and with reference to that of BV at 5 minutes yields by interpolation:

From Table IV, BV at 5 minutes = R at 36.8 minutes
" " V, BV at 5 minutes = R at 39.0 minutes

Av. = 37.9 minutes
or BV = 7.6 R in terms of time of exposure required.

In Tables IV and V are given also a few evaluations for exposures to red at 50 cm. for five minutes. When the average of these is compared with the average of the exposures at 150 cm. for forty-five minutes, the latter is less than the former, indicating the possibility that diminution of intensity is not compensated for by a corresponding increase in the length of exposure. In Table V are given a few exposures to yellow-green, which, when taken with the evidence furnished by the section-method, indicate that the efficiency of this region of the spectrum is not greater than that of the blue-violet and that, although they rank close together, it is probably less.

IV. Discussion.

A. METHODS AND CHECKS EMPLOYED.

A comparison of the two methods employed in the foregoing investigation will show how they supplemented and confirmed each other.

The first procedure, by which the eye was sectioned and studied microscopically, had various limitations as compared with the second: — the delay necessitated by the removal of the cuticula; the use of only one color for each animal; and ignorance of the initial position of the pigment. From the last named limitation as a premise, it could be argued that the final differences obtained in the effects of the various colors might be due to initial differences in the position of the pigment. At the season of the year, however, when the section method was applied, viz. during May and June, the crayfish were in vigorous condition, and since animals of the same sex and as nearly the same size as possible were selected for a given series, and since the check eyes, not only of Table I but also of earlier preliminary tests, gave satisfactory evidence that the six hours allowed for dark-adaptation was sufficient, the error from that source was probably very slight. On the other hand, sections offered greater precision for the determination of the photokinetic effects, in that they showed the actual distance traversed by the pigment. This was not a uniform amount over the whole retina. The maximum migration occurred at the center, caused probably by the concentration of light in the vertical image of the Nernst filaments formed on the retina, while on either side the amount gradually diminished. This diffusion effect is shown in Plate **3**, Fig. 6. In the left half of the photograph the emigrated pigment has been indicated, by retouching the photographic print with ink, as a darker band in order to distinguish it from the tapetum, which also photographed dark. At the center they practically coincide in extent, but at the extreme left the pigment band lies proximal to the outer limit of the tapetum. The degree of gradation away from the center was often an aid in distinguishing the influence of the different colors, because it was most abrupt for the red and least so for the blue-violet.

Direct observation of the photokinetic changes supplemented the section-method in the following respects: the same animal could be tested for all the colors; a larger number of observations could be made, on account of greater facility; the unexposed eye served as a check upon the other; and, since the glow could be determined by the colored light at the beginning of the exposure, the same initial condition of the eye could be ensured for each test. The chief objection to the method lay in the fact that it involved a judgment by eye and an artificial method of recording the amount of migration. Since each record was made independently of previous ones for the same animal, and since as a rule the large number of animals used at one time prevented the retention in ones memory of the relative values of previous exposures, the chance for being mentally biased in recording observations was practically eliminated. After a little practice the repetition of certain trifling peculiarities of individual eyes in the records gave ground for the belief that a certain degree of accuracy had been acquired in judging and recording conditions. In order to establish it beyond a doubt, however, a check was made in the following manner: — eleven eyes, eight in one series and three in another, were sectioned for the purpose of comparing the actual amounts of migration with the conditions as observed immediately after exposure and recorded after the animals had been plunged into hot water. In my own estimation the gradation of the migration in the sections practically coincided with the gradation as shown by the records; but in order that an unbiased comparison might be made, Professor Parker also arranged the slides and the records independently of each other in the order of progressive migration. In Table VI are given our respective comparisons of the sections and records for the two series of eyes. In the first series there was no doubt about the minimum condition in eye No. 131, the maximum in 136 and an intermediate condition in 134. In my judgment 133 and 138 also 135 and 137 were so close as to be interchangeable. In series II there was little uncertainty about the correspondence of sections and records. This series is reproduced in Plates **4** and **5**, Figs. 7a, 7b, 8a, 8b, 9a, and 9b, and shows the recorded observations of the glow together with a photograph of a section of eye 140, 139, and 141, respectively. These comparisons of the recorded estimated condition with the actual condition in sections gave fair evidence that the method was a legitimate one and that the results obtained by it were valid within an error of 10–15 percent.

TABLE VI.

Observation-records checked by sections of the same eyes. The horizontal lines P and D indicate the arrangement of both sections and records in the order of progressive amounts of migration (left to right) made by Professor Parker (P) and myself (D), respectively. The numbers are those by which the different eyes were designated.

I.	P	Sections	131	133	138	134	135	132	137	136
		Records	131	138	133	132	135	134	137	136
	D	Sections	131	133	138	132	134	135	137	136
		Records	131	138	133	132	134	135	137	136
II.	P and D	Sections	140	139	141					
		Records	140	139	141					

My experience with the two methods and in working with the animals at different seasons of the year brought out a fact about the migration, various aspects of which had been noticed before by Exner ('91), Fick ('91), Parker ('97), and others, and this must enter into the final consideration of the results. Exner had observed that the rate of the migration diminished as the animals became feeble, and stopped altogether as they approached death. Fick discovered in the frog that there was a latent period between the initial stimulus by the light and the inception of the migration, *e. g.* frogs exposed from two to four minutes to light and killed showed no migration, whereas others exposed for the same length of time and then put in the dark for twenty minutes exhibited some. In my own experiments both of these peculiarities appeared and were found to be correlated. In the spring when the animals were vigorous, the pigment responded with great celerity in the first minute or two of exposure and progressed at a diminishing rate thereafter; but in the winter a stronger intensity and longer exposure were needed to elicit the migration. From this it was concluded that the latent period was a direct function of the physiological state of the animal.

The question of this latent period came up in connection with those experiments in which the time of exposure was varied while the intensity remained constant. Since there was a manifestation of inertia in the form of initial latency, might not there also be another manifestation of it at the close of the exposure in the form of momentum? Would the pigment, after a five-minute exposure to blue-violet or after the longer exposures to red, continue to migrate upon cessation of the stimulus? If there had been any appreciable after-effect of the exposure, it would have complicated the comparison of the effects of the two colors; but tests made by exposing to red at 50 cm. for five minutes then restoring the animal to the dark for ten, fifteen, thirty, or forty-five minutes, yielded no detectable increase in the migration.

A further source of error might have come from the apparatus. If there were any leakage of white light, its additional effect on the migration would have been much greater in the long exposures to red than in the short one to blue-violet. Although diaphragms had been interposed (Fig. F) to eliminate such an error, a check was employed which settled the question conclusively. A series of six independent exposures upon a single photographic plate (Seed's "Gilt Edge 27") was made as follows:— at 50 cm. for periods of thirty, sixty and ninety seconds; and at 150 cm. for periods of 270, 540 and 810 seconds, the last three periods being respectively nine times as long as the first three. The results are reproduced in Plate 5, Figs. 10 *a–f*, and are according to the above order. The exposures in each vertical pair (e. g., *a* and *d*) are comparable, the one above being an exposure to a strong intensity for a short period, and the one below to a weak intensity for a compensatingly long period. The difference in the actinic effect of corresponding exposures was almost imperceptible. The series was made with the diaphragms in position as during the experiments with the animals. A test with the diaphragms left out yielded the result that the long exposures at 150 cm. showed a greater actinic effect than the corresponding ones at 50 cm. Such a delicate test, therefore, proved not only the freedom of my results from any error due to leakage of light, but also the efficiency with which diaphragms may be used to exclude extraneous, reflected light in experiments of a similar nature.

In order to make sure that the equality of intensity was not disturbed by differences in the rate at which the several colors diverged after issuing from the light-box, I measured the width of each band at two positions. The ratios which obtained between widths of the color-bands at one position were found to hold also at the other, so that the intensities were consequently always the same.

The most important checks throughout the investigation have been:— the radiomicrometer for intensity; check animals for the series run by the section method; the unexposed eye of each animal for the series of direct observations; the verification of the records of direct observations by sections; and the photographic test for leakage of white light.

B. Summary of Observations.

1. Different regions of the spectrum at equal intensity elicited different amounts of pigment migration.
2. Blue-violet was more potent than red, as evidenced both by sections and by direct observations of the crayfish eye.
3. The quantitative expression for the difference in efficiency as determined by varying the intensity of the light was $BV = 5.6\ R$; and
4. As determined by varying the period of exposure, $BV = 7.4\ R$.
5. Blue-violet, green, and yellow ranked close together, but of the three blue-violet was probably the more efficient in evoking the migration.
6. The rate of migration of pigment varied with the physiological condition, being slow in a feeble or sluggish animal and more rapid in a vigorous one.
7. The quantitative expression for the efficiency of blue-violet in terms of that for red was probably a variable dependent on the physical condition of the animal.
8. A bleaching of color from metallic orange to red and then to dull yellow was observed in the eye exposed to light. The possibility is that this phenomenon is due to a chromatic substance located in the rhabdomes, which undergoes a partial bleaching and is analogous to visual purple in the vertebrate retina.

C. The Function of the Pigment.

The fact that the migration of pigment varies with the color of the light evoking it has an important bearing upon the problem of the function of the pigment. Hesse (:02) has reviewed the early conceptions of its function prior to the knowledge of the photokinetic phenomenon, has discussed the modern theories about it, and has added new light on the subject from his own investigations.

Hesse's anatomical study of the optic organs of many invertebrates yielded two facts bearing upon the present problem: — (1) in certain leeches and annelids are found unpigmented cells which, because of their resemblance to pigmented cells or eye-spots in close relatives, are undoubtedly visual cells; (2) often where pigment is present it is either separated from the sensory cells by an intervening tapetum, as in Pecten and some insects, or it is diversely situated in eyes of closely related species, as among the gastropods. From this it is seen that pigment is not essential to the physiological reception of light nor — as the experiments of Harrington and Leaming (:00) and of Mast (:11) on Amoeba indicate,— even of color; and although pigment be present, yet its loose association with the visual cell bars attributing to it a primary rôle. When the relation between the two is closer, the pigment-cup, by partially screening the cell, enables the animal to determine the direction whence the light comes.

The phenomenon of migration of the pigment probably arose with the evolution of the eidoscopic eye, with which, at first, only moving objects, and later stationary forms as such, were perceived by the animal. At the outset, before the significance of the pigment can be logically discussed, an answer must be sought to the fundamental question: — where are the receptive organs in the eye of the crayfish located?

Parker ('95) has emphasized two essential features which the receptive organ must have:— first, a dioptric mechanism for transmitting the light to it; and, secondly, nerves for conducting the stimulus to the brain. The phenomenon of the pigment-migration may aid in identifying the receptive region. Thus, Hesse (:00) has questioned Grenacher's conclusion that the rhabdome in the retina of the cephalopod is the sensory organ, and has pointed to another structure as being more probably the sensory organ, because of its closer relation to the emigrated pigment. In the crayfish the optical, neurological, and photokinetic evidence — for a discussion of which see Parker ('95) and Hesse (:01) — points to the rhabdome as the region in question. As to the vertebrate eye, the evidence of neurofibrillae on the outer segments of the rods and cones furnished by Hesse (:04) and Howard (:08); the observations by Van Genderen Stort ('87), Chiarini (:04 and :06), and Herzog (:05) of the pigment-migration extending only out to the ellipsoids of the rods; the discovery by Birnbacher ('94) that the ellipsoids stain deeply in a dark-eye, but poorly in a light-eye with an acid stain; and the observations by Hess (:07 and :10) of a shortening of the range of perception in the blue end of the spectrum among birds and reptiles due to the colored oil-drops in the ellipsoids of the cones, all go to indicate that the outer segments of the rods and cones together with the ellipsoids are probably the receptive organs.

Since the pigment migrates outward to surround the rhabdomes in arthropods and the outer segments of the rods and cones in vertebrates, it evidently plays some rôle in the vision of the animal.

Early investigators were tempted to attribute to it a primary function, that of transforming the light-energy into a form appropriate for stimulating the nerve-endings on the receptive organs. Kühne ('78) suggested that this might be by mechanical friction of the pigment-needles on the rods and cones, or perhaps by the end-products of chemical decomposition of the pigment under the influence of light. No evidence has been found for the mechanical view. As to the second view, support might be found in the following facts: — Kühne ('78) succeeded in bleaching pigment extracted from the eye and exposed to sunlight for several weeks; Stefanowska ('90) observed that in some insects (e. g., Eristalis tenax) the pigment was resolved into oil-drops, while Chiarini (:04) found that in many animals it diminished in quantity under the influence of light; and Raehlmann (:07) beheld under the ultramicroscope an actual bleaching of granules in the processes of the pigment-cell. Kühne, however, never obtained bleaching in the living eye, and that which Raehlmann observed was in granules other than of pigment. Furthermore, since we know that pigment is unnecessary for the perception of light or color in many invertebrates, according to the observations of Hesse (:01), Mast (:11) and others, and likewise in vertebrate albinos, further that the rate of migration is relatively slow, and that vision is good after the migration has ceased, the theory ascribing to the pigment a primary rôle of chemically stimulating the visual cells seems hardly tenable.

Another theory attributes to the pigment the quasi-nutritive function of replenishing the exhausted visual cells. Kühne found that an excised eye of a frog with the pigment-epithelium intact regenerated the visual red, while a retina from which this epithelium had been removed did not. Chiarini (:06) believed that the migration was due to chemotropism and that the pigment thereby replenished the rods and cones, whose substances had been exhausted by the influence of light. Raehlmann (:07) thought that the pigment was a vehicle for conveying visual red to the rods. According to the observations and deductions of these investigations the pigment would therefore assume a secondary rôle, while visual purple, or some other substance in the photoreceptive elements, would become the chief factor in transforming the light-energy into the appropriate stimuli.

The theory now most generally accepted (by Stefanowska '90, Szczawinska '90, Exner '91, Parker '95 and '99, Hesse :02, Garten :07, and Doniselli :07) is that the pigment is a protective mechanism for regulating the amount of light entering the receptive organ, and for rendering better definition to the image by preventing irradiation.

This, however, still leaves open the question as to how the pigment migrates. What are the mechanics of the phenomenon? This problem may be resolved into two questions:— first, whether or not the migration is under the control of the central nervous system; secondly, whether the migration is due to an intra- or to an extracellular activating force.

As to the first question, Engelmann ('84), Fick ('90), Kiesel ('94), Herzog (:05) together with Nahmacher, Lodato, and Pirrone (cited by Herzog) have obtained affirmative evidence, whereas the investigations of Hamburger ('88), Fick ('91), Parker ('97), and v. Frisch (:08), on the contrary, have indicated that the migration is independent of the central nervous system. Since some of the evidence is convincing for both views, it is possible that the two views are reconcilable on the assumption that the migration of the pigment is induced primarily by the direct stimulus of light, while the central nervous system exerts a secondary influence upon it, perhaps of slight inhibition or of occasional stimulation.

The evidence both for an intra- and an extra-cellular cause of the migration I shall consider with respect, first, to the direct response of the pigment, and secondly, to the indirect response through a reflex.

1. Direct response of the pigment. (a) *Evidence for an intracellular activating force.*— Kühne ('78) observed that after injecting pigment into the blood of a salamander those white corpuscles which had ingested a few particles of pigment were more kinetic under the influence of light than the others, which had not. Herzog (:05) believed that the pigment absorbed heat and emitted it in the form of either kinetic or chemical energy, which might stimulate the receptors or else might be but an indifferent form into which the radiant energy had been diverted. This view, according to which the migration would be proportional to the heat-energy absorbed by the pigment, is contradicted by the results obtained with colored

lights of equal intensity. The phenomenon of an electric current induced in the eye of a frog by light, studied by Gotch (:03 and :04), might offer a basis for the interpretation of the migration. If there were a difference in potential produced in the pigment-cell it would not be one comparable, however, to that set up in the radio-micrometer proportional to the radiant energy absorbed, but rather to that created by some chemical change in the cytoplasm. Von Frisch (:08), however, obtained no pigment migration in the eye of a shrimp upon applying electrodes to it.

(b) *Evidence for an extracellular or chemotropic force.*[1] In many insects and crustaceans there is a migration of distal pigment inward and of proximal pigment outward, but in both cases toward the rhabdome. In the vertebrate eye the migration is outward; the pigment surrounds the outer segments of the rods and cones, and stops pretty definitely in the vicinity of the ellipsoids of the rods. The ellipsoids of both elements stain less deeply with acid eosin in the light-eye than in the dark-eye, indicating that a chemical change from an alkaline to an acid condition has been produced by the light. As Birnbacher ('94) pointed out, however, this condition in the ellipsoids might be but the end of a series of changes which have gone on in other parts of the rods and cones. Thus substances, chromatically visible or otherwise, in the outer segments might also be concerned in the chemotropism.

2. INDIRECT RESPONSE THROUGH A REFLEX. Since Fick ('90) and Kiesel ('94) observed in the frog and moth respectively a migration in the dark, it would seem that the optic nerve has a motor function. Although I do not know whether there are any nerves connecting with the pigment-epithelial cells in the vertebrate eye through which the motor impulse might be dispatched, in the arthopod, according to Parker ('95) and Hesse (:01), neurofibrillae pass up through the substance of the proximal retinular cells and into the rhabdome. Whether the distal retinular cells are likewise innervated, I do not know. Such nerve-connections would mean an intracellular stimulus to migration in the dark as opposed to an extracellular chemotropic stimulus to migration in the light. Even if the migration in the light were evoked through a reflex, there might be a causal relation between the chemical change evident in the retina and the initiation of this

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[1] By chemotropic force I mean an attraction-force (whether due to a change in electric potential, osmotic pressure, affinity of one chemical substance for another, or similar process) occasioned by the dissociation of some chemical substance in the receptive organ under the influence of light.

reflex; so that whether the chemical change acted upon the pigment directly through chemotropism or indirectly through a reflex, the amount of migration would nevertheless be proportional to the chemical reaction. This interpretation is quite in keeping with the known facts:— first, that at equal intensity blue-violet is vastly more potent than red actinically (about 20,000 times according to a photographic test which I made upon a Seed's "Gilt Edge 27" plate — true however only for the given emulsion and given intensity); secondly, that blue-violet is more efficient than red in evoking the migration of pigment.

The final explanation of the pigment as a protective mechanism would be, according to the above deductions, that it is correlated with the sensitivity of the receptive organs to those wave-lengths which stimulate them to the greatest chemical activity. Since the data from which the deductions have been made referred partly to the compound eye and partly to the vertebrate retina, and since the quantitative statement for the migration of pigment in both cases was not very large in amount, and since, further, the facts were often conflicting, the above conclusion is but a tentative one.

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Explanation of Plates.

Plate **1** exhibits the typical extreme condition for an eye adapted to light (Fig. 1), and for one adapted to darkness (Fig. 2), both as seen by direct observation (Figs. 1a, 2a) and in a section of the eye (Figs. 1b, 2b).

Plates **2** and **3** show sections of four eyes exposed respectively to blue-violet, green, yellow, and red of equal intensities and for the same length of time. Evaluations for the relative amounts of migration are given in Table I, series 3.

Plate **4** and Fig. 9, Plate **5**, are checks on the migration of pigment as recorded by direct observation (a), and as revealed by sections of the same eyes (b).

Plate **5**, Fig. 10 is a photographic test, which gives evidence of no error due to leakage of white light from the light-box.

Figs. 1a, 2a, 7a, 8a and 9a are copies of the original records. The magnification is about 10 diam. The orange (metallic) glow of the eye (Fig. 2a) was due to light reflected by the tapetum back through the orange-colored rhabdomes. Bleaching to a dull yellow occurred in the light (Fig. 9a). Concomitant with this a dark area appeared, which was an index of the amount of pigment-migration: in Fig. 2a there is none; Figs. 7a, 8a and 9a show successive steps of the change induced by light, which culminates in Fig. 1a. The appearance of the dark area in only the upper half of Figs. 7a, 8a, and 9a was probably an optical effect due to refraction in the cones.

The figures of all the sections, which were median longitudinal ones cut in an antero-posterior direction, are photographic reproductions made with a combination of Leitz oc. 4 and Zeiss a^* obj. upon a Seed's 26 plate and magnified about 40 diam.

PLATE 1.

Fig. 1a. Standard *light-eye*: diagrammatic representation of external appearance in daytime showing pseudopupilla.

Fig. 1b. Standard light-eye: exposed 6 hr. to light of north window; photograph of median section, × 40. A single ommatidium sketched in left half:

bm,	basement membrane	*dp,*	distal retinular cells
c,	cone	*pp,*	proximal " "
cr,	corneal hypodermis	*rh,*	rhabdome
ct,	cuticula	*t,*	tapetum

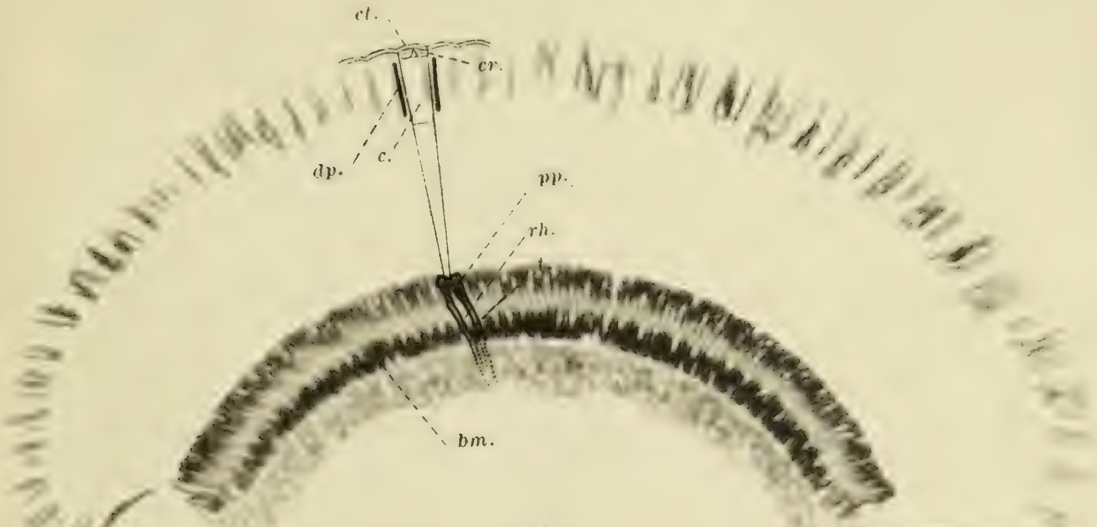
Pigment of the proximal cells has migrated from the region below the basement membrane and has become concentrated around the nuclei in the tips of the cells.

Fig. 2a. Standard *dark-eye*; diagrammatic representation of eye after 6 hr. in the dark when examined with a flash-light.

Fig. 2b. Standard dark-eye: adapted to dark for 6 hr. and killed in the dark; photograph of median section.

npp,	nuclei of proximal retinular cells (drawn in ink).
rp,	pigment " " " "

Retinal pigment lies mostly behind the basement membrane. The left half of the photograph is retouched to show the blunt processes of pigment of the proximal retinular cells distal to the basement membrane, which were not differentiated from the tapetal pigment by the camera.


ct.
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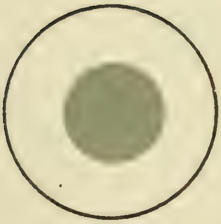
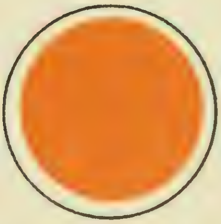
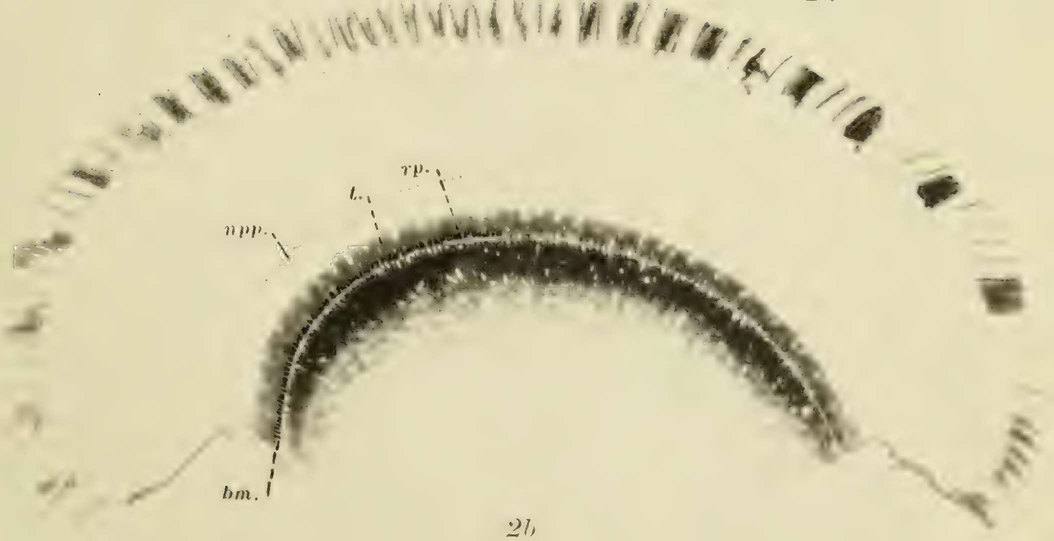
1b



1a



2a



2b

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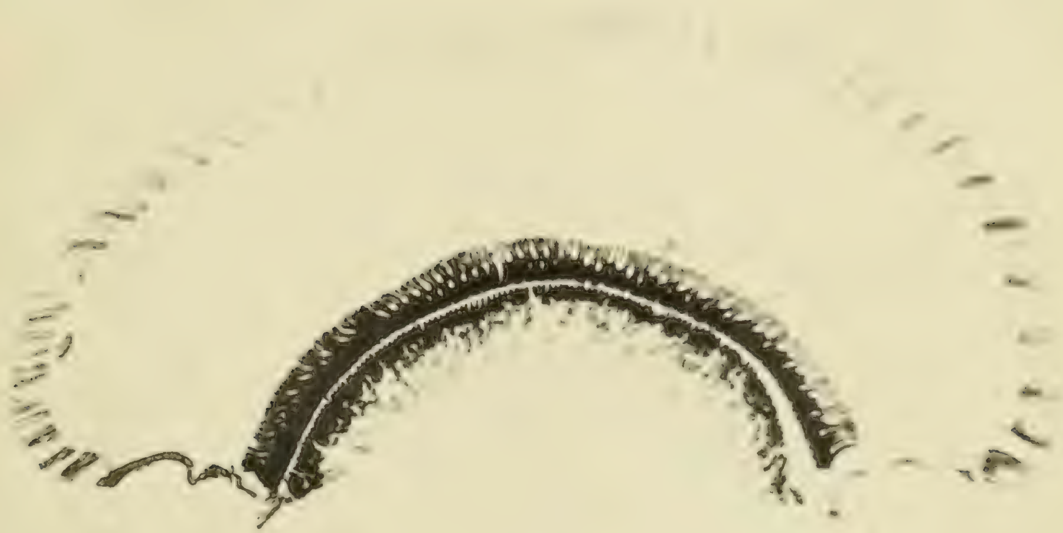
PLATE 2.

Fig. 3. Photograph of a median section of an eye exposed to *blue-violet* at a distance of 550 cm. from the light-box for 1 min., after having been 6 hr. in the dark. × 36. The pigment reaches to the nuclei of the retinal cells.

Fig. 4. Photograph of a median section of an eye exposed to *green* at 550 cm. for 1 min., after having been 6 hr. in the dark. × 40. The pigment has migrated beyond the tapetum (indicated by broken line in left half of the figure), but not out to the nuclei.



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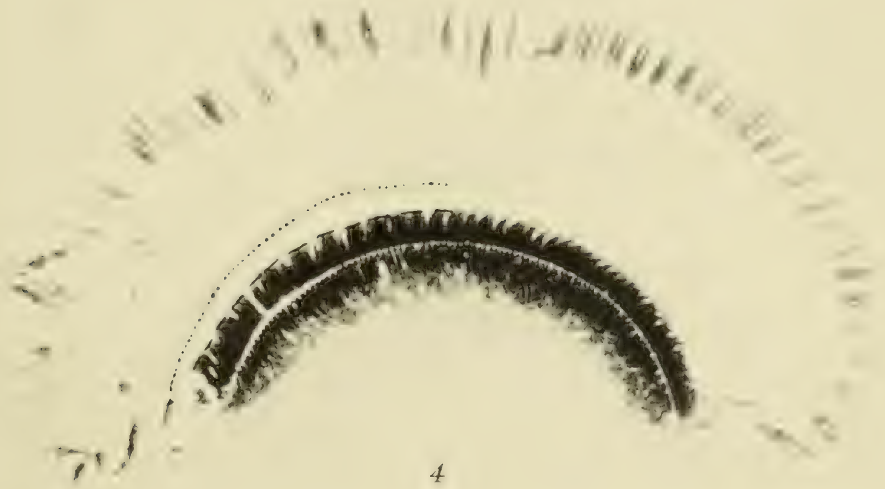

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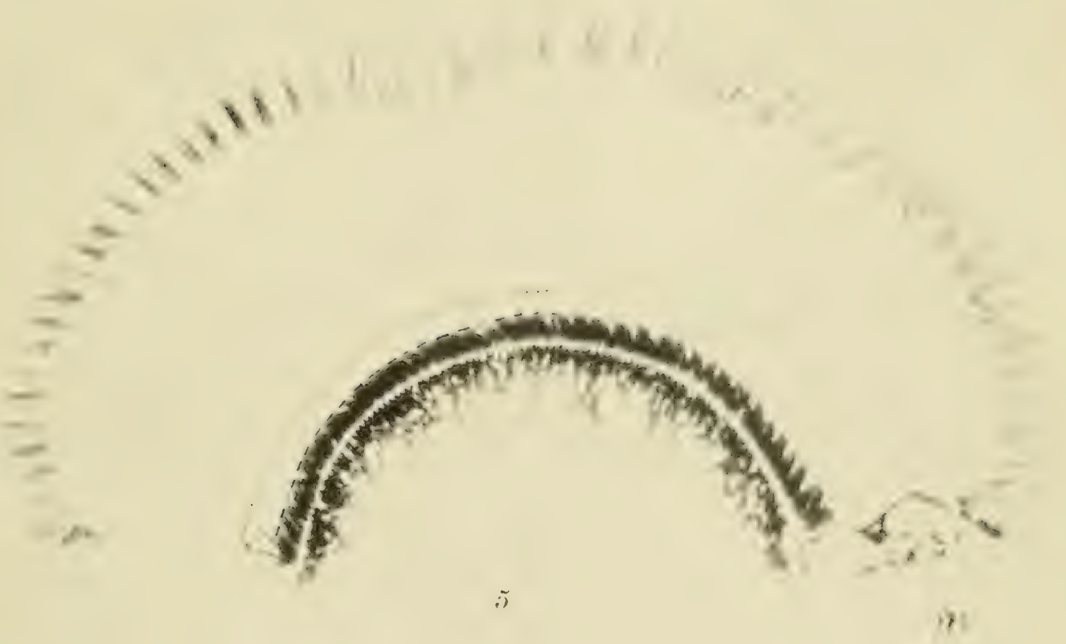
PLATE 3.

Fig. 5. Photograph of a median section of an eye exposed to *yellow* at 550 cm. for 1 min., after having been 6 hr. in dark. × 40. The pigment is coextensive with the tapetum (to the broken line in the left half of the figure).

Fig. 6. Photograph of a median section of an eye exposed to *red* at 550 cm. for 1 min., after having been 6 hr. in the dark. × 45. The pigment is coextensive with the tapetum only about the center; on the sides (as indicated by the diminishing width of the dark band sketched into the left half) it falls short of the extent of the tapetum.



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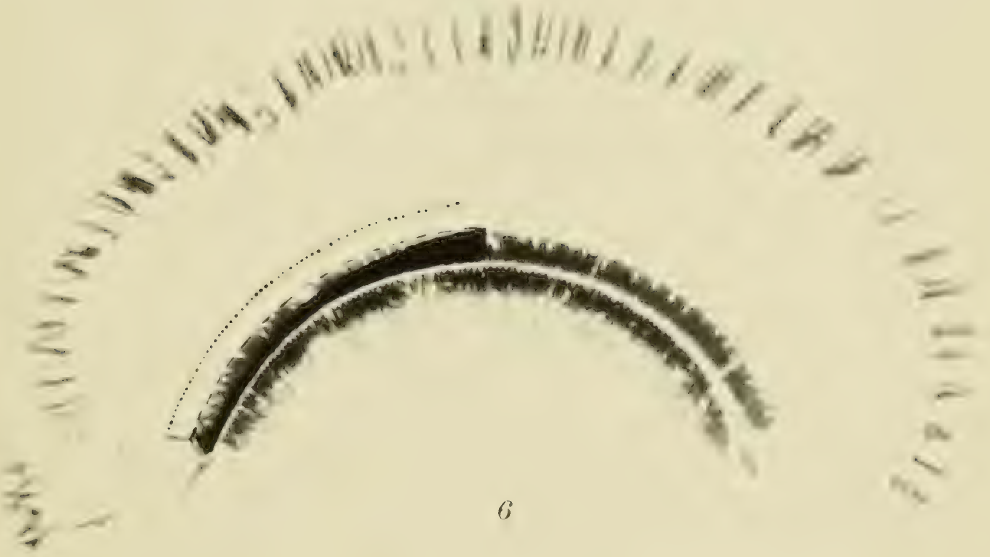

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PLATE 4.

Figs. 7a, 8a, 9a are copies of the original diagrammatic records of eyes 140, 139, and 141, respectively.

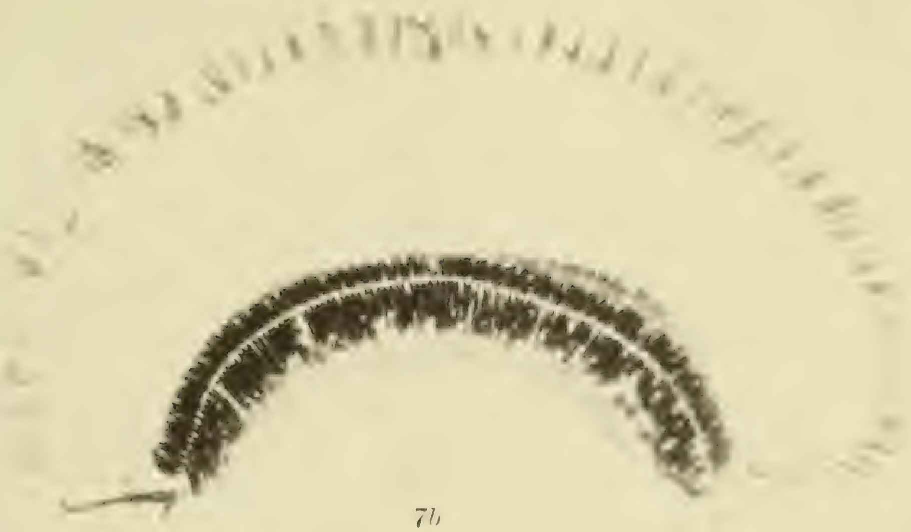
Figs. 7b, 8b, 9b are photographs of sections of the same eyes as those exhibited in Figs. 7a, 8a, and 9a after they had been killed, to show the changes induced in them by exposure to light.

Fig. 7a. Observation record of eye 140, dark 6 hr. Color, ruddy orange. Slight darkening in upper half.

Fig. 7b. Section of eye 140, × 43. On the left the pigment extends slightly beyond tapetum, on the right there is a heavier local migration.

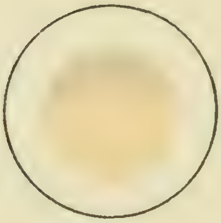
Fig. 8a. Observation record of eye 139, dark 6 hr. Color, yellowish orange. More darkened in upper half than in Fig. 7a.

Fig. 8b. Section of eye 139, × 42. The pigment extends in a more definite fringe beyond the tapetum than in Fig. 7b; a heavy local migration is observable on the left.

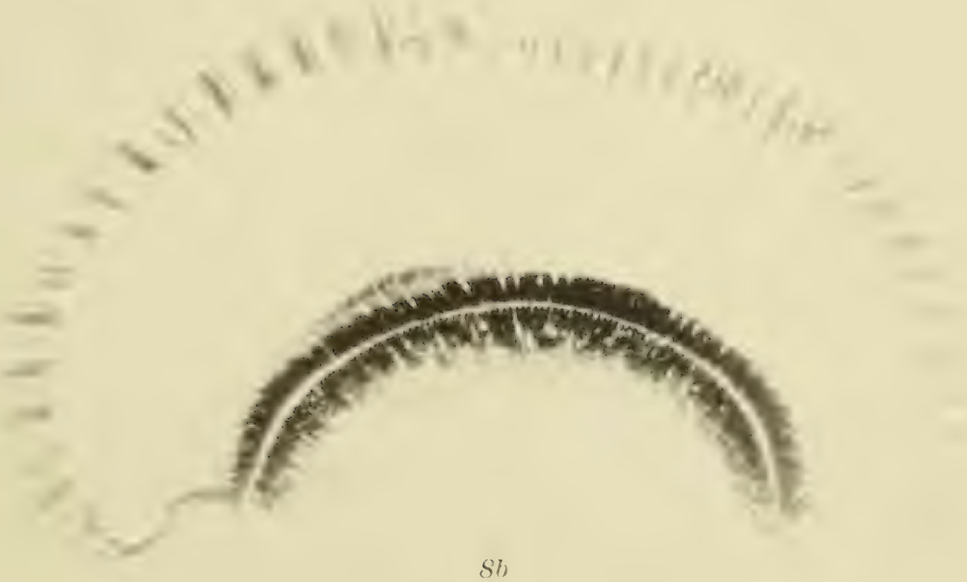


7b

7a



8a



8b

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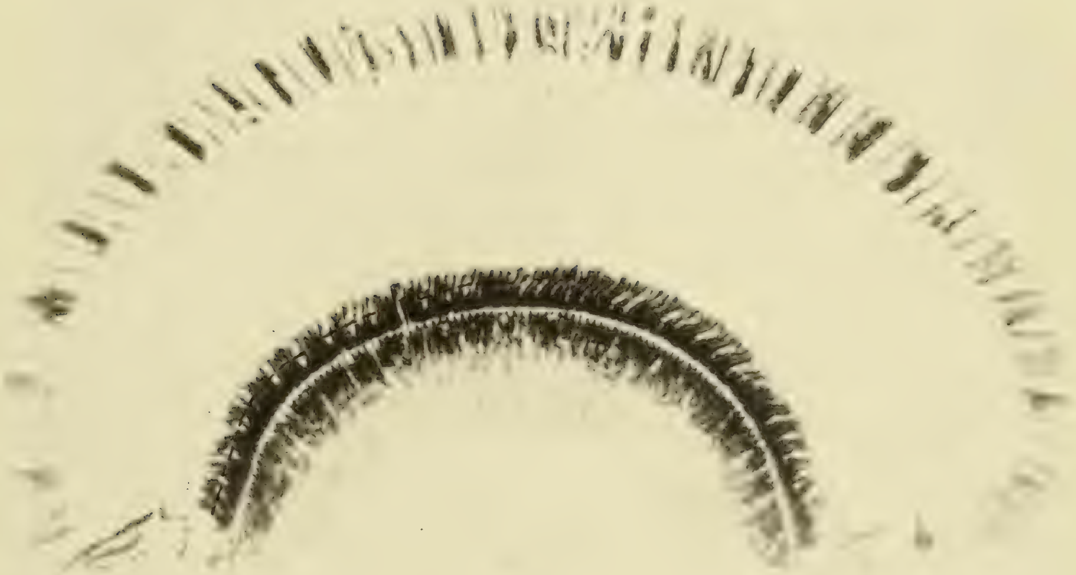
PLATE 5.

Fig. 9a. Observation record of eye 141, dark 6 hr. Color, yellowish orange. Pronounced darkening in the upper half.

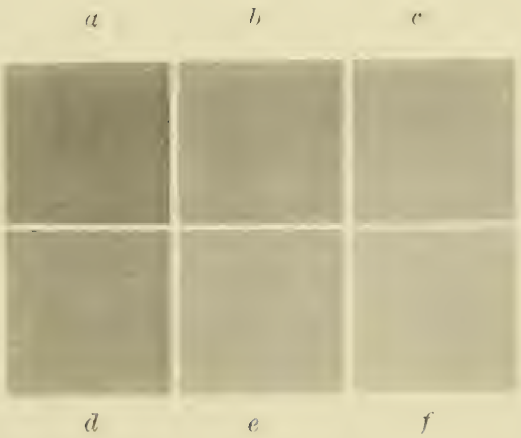
Fig. 9b. Section of eye 141, × 41. The extent of the migration is about twice the width of the tapetal layer, but not out to the nuclei.

Fig. 10. Photographic test of light-box for leakage of white light. A Seed's "Gilt Edge 27" plate, substituted for the animal as in text-fig. F, had six separate exposures made on it as follows: At 50 cm.: a, 30 sec.; b, 60 sec.; c, 90 sec. At 150 cm.: d, 270 sec.; e, 540 sec.; f. 810 sec.

9a



9b



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Bulletin of the Museum of Comparative Zoölogy
AT HARVARD COLLEGE.
Vol. LIII. No. 7.

EFFECTS OF RADIUM ON LIVING SUBSTANCE.

I. THE INFLUENCE OF RADIATIONS OF RADIUM UPON THE EMBRYONIC GROWTH OF THE POMACE-FLY DROSOPHILA AMPELOPHILA, AND UPON THE REGENERATION OF THE HYDROID TUBULARIA CROCEA.

By E. D. Congdon.

CAMBRIDGE, MASS., U. S. A.:
PRINTED FOR THE MUSEUM.
February, 1912.

No. 7.—CONTRIBUTIONS FROM THE ZOÖLOGICAL LABORATORY OF THE MUSEUM OF COMPARATIVE ZOÖLOGY AT HARVARD COLLEGE, UNDER THE DIRECTION OF E. L. MARK, No. 228.

Effects of radium on living substance.— I. *The influence of radiations of radium upon the embryonic growth of the pomace-fly Drosophila ampelophila, and upon the regeneration of the hydroid Tubularia crocea.*

By E. D. Congdon.

Introduction.

These studies on the effects of radium, the first two of which are here published, were begun in 1906, at the suggestion of Professor Mark. The work was carried on under his direction in the Zoölogical Laboratory at the Museum and in conference with Prof. Theodore Lyman of the Jefferson Physical Laboratory. The results on other parts of the work are reserved for further experimentation before publication.

Space does not permit here an extended description of the three types of radiation given off by radium, the alpha, beta, and gamma rays. According to the commonly accepted corpuscular theory of matter, two of the three, the alpha and the beta, consist of particles of matter. The third, the gamma rays, are movements of the ether. The alpha rays are so little penetrating that a few sheets of paper will absorb them; the beta rays, on the contrary, are more penetrating and may even traverse a considerable thickness of metal. The gamma radiations exceed the beta rays, as well as X-rays, in their penetrating power. The larger portion of the energy emitted by unscreened radium is in the form of alpha rays. Further information as to radium and its radiations may be found in such standard works as Rutherford's Radioactivity and his Radioactive Transformations.

In spite of the numerous investigations on the biological action of the three types of radium rays which have appeared since the early notices from Becquerel and others, not much progress has been made in analyzing their action. This is evident from the fact that we know almost nothing of their distinguishing biological effects otherwise than those arising from differences in their penetrating power. An explanation of the lack of success in determining their differences of biological action is to be found in the difficulties encountered in isolating and measuring the radiations. Furthermore if a pure radiation of known intensity be obtained, the changes as to intensity and quality of all three types of radiation in their passage through the tissue remains to be reckoned with.

Drosophila.

In the following experiments with the eggs of Drosophila the quality and intensity of the alpha, beta, and gamma radiations are known only approximately; but the complication due to the change of character of the radiations at different depths from the exposed surface of the egg is largely eliminated by the minuteness of the egg (less than 0.1 mm. in diameter) and the thinness of the egg case.

In most exposures a hundred milligrams of impure radium of one

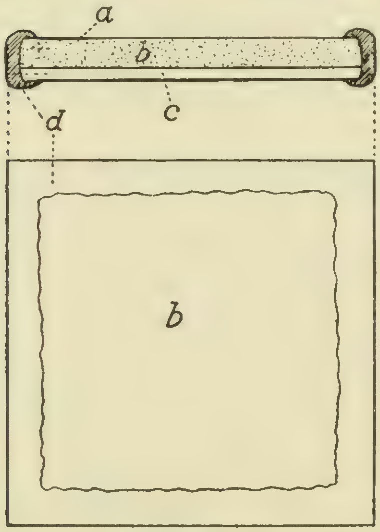

Fig. 2.

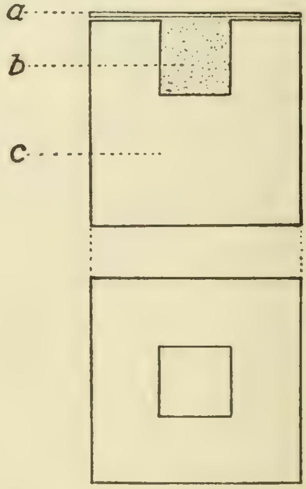

Fig. 1.

Fig. 1. Diagram of lead cell used to hold the radium. The upper portion of the figure is a vertical section; the lower a horizontal section as the level *b*. Magnified one and one third diameters.

a. Paper screen, two thicknesses of black paper such as is used to protect photographic plates; *b*, radium; *c*, lead cell.

Fig. 2. Diagram showing construction of the cell used to contain the radium (the weaker sample) in most of the experiments except those on the eggs of Drosophila. Magnified one and one half diameters.

a. Mica roof of cell; *b*, Radium; *c*, Glass floor of cell; *d*, sealing-wax rim.

thousandth the strength of the pure bromide was the source of the radiations.[1] It was contained in a cubical cell of lead (Fig. 1) the walls of which were 5 mm. thick. To eliminate so far as possible other influences than those of the radium, the experiments were conducted in a constant-temperature chamber (Fig. 3), and the eggs were kept moist by an irrigating device (compare Fig. 4). The eggs were supported above the cell on a sheet of filter paper parallel to the surface of the radium. The intensity of the beta radiations was controlled by varying the space between radium and eggs. By multiplying the number expressing decrease of intensity due to the spreading of the radiations (based on radiographs at different distances above the radium) by the factor for absorption of beta radiations in the air,[2] quantities were obtained expressing the relative strength of treatment in the different experiments. The length of exposure was twenty-four hours. The decrease of penetrating power of the radiations at the different heights due to absorption by the air was negligible.

The material for a single exposure consisted of from two hundred to four hundred fly eggs, which were removed to the centers of two small moist pieces of filter paper, an equal number on each piece, within two hours after being laid. The filter paper bearing one set was supported on a light wooden frame (carrier) at the desired distance from the radium (*b*, Fig. 4); the other, protected from the radiations by a mass of lead two inches thick, was kept in the same thermostat in which the exposure was going on. An automatic irrigating device (Fig. 4, *d*) prevented the eggs from drying. The larvae began to come out of the egg cases soon after removal and were usually nearly all hatched in twenty-four hours. The average periods of incubation for the exposed and control eggs were never so different that the two sets did not well overlap in time of hatching. A comparison of the number of eggs that hatched out from the exposed box during the period of the experiment with the number that hatched from the control during the same period, afforded a basis for an approximate expression of the rate of growth of irradiated eggs. The comparison was made at the time when approximately half of the controls were hatched. It may be objected that time of hatching does not necessarily express amount of growth, and has to do only with the casting off of the egg coat itself. Such an explanation is

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[1] Thanks are due to Mr. Hugo Lieber and Dr. Theodore Lyman for the loan of radium.

[2] Rutherford, E., Radioactivity. University Press, Cambridge, 1905, xi + 580 pp.

Fig. 3.

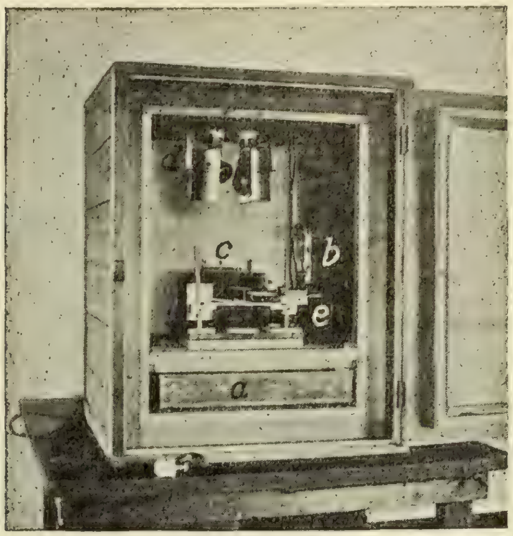

Fig. 4.

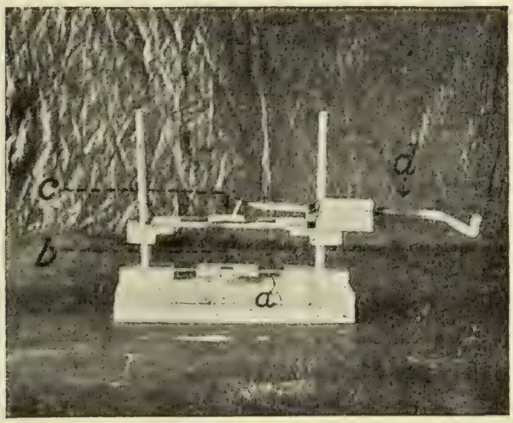

Fig. 3. Interior view of constant-temperature chamber with double walls, the double-glazed glass door open. This cubical chamber measured about twenty inches to the side, and contained, below a removable wooden grating, a copper tank (*a*) filled with water heated by an electric coil regulated by an automatic device (*b*) depending on the expansion of a curved metal strip for making and breaking the circuit. In very cold weather this heating tank was supplemented by a pair of 32-candle-power electric lamps (*d*) with blackened bulbs suspended from the middle of the roof of the chamber and partially enclosed in screens to prevent local and unequal heating of the chamber. Resting on the floor of the chamber above the tank was a thermograph (*c*), and the support (*e*) for radium, eggs, and a small reservoir for moistening the eggs.

Fig. 4. Enlarged view of the support (*e*, Figure 3), showing the base (*a*), on which rests the lead cell (*b*) containing the radium, the two cylindrical posts supporting the movable carrier for the eggs (*c*), and a movable glass-tube water reservoir (*d*) terminating at one end in a fine nozzle, from which a plug of fine threads leads to the moist filter paper supporting the eggs. The adjustable egg-carrier allows one to regulate the distance between eggs and radium.

very unlikely because the radiations give up their energy not only to the surface of the egg, but to the internal portions as well. Abundant analogous experiments with light on insect eggs, larvae, and pupae have given accelerations or retardations of growth.

TABLE I.

Number of the experiment.	Intensity of beta rays.	Percentage of retardation.	Average.
1	48	52	34
2	48	16	
3	48	34	
4	38	18	29.6
5	38	35	
6	38	51	
7	38	15	
8	38	29	
Averages	41.7	31.2	

Table I shows the effect upon growth inside the egg case due to exposure to the one hundred milligrams of impure radium at the distance one centimeter (nos. 1–3) or two and a half centimeters (nos. 4–8). The intensities were calculated by the method already described. Alpha rays were screened off by using just sufficient paper to absorb them all. The beta radiations under these conditions contained about three fourths the energy of the remaining (beta and gamma) radiations. The other fourth, consisting of gamma radiations, must have given up to the eggs, relative to its energy content, much less than the beta radiations, because the former are the more penetrating. The effect of either intensity of exposure was a retardation. The average of the retardations produced by intensity 48 was greater than the average for the intensity 38, i. e. the retardation was greater with the more intense exposure.

TABLE II.

Number of the experiment.	Intensity of beta rays.	Percentage of retardation.	Percentage of acceleration.
1	33	14	
2	31	20	
3	25		2
4	23	2	
5	20	4	
6	15		4
Averages	25.5	6.7	1.0

In Table II, containing exposures at distances up to five centimeters from the radium, it is shown that at most only small retardations have occurred and that sometimes there was an actual acceleration. The two tables, then, show a proportion between the intensity of exposure and the effect. A treatment with an intensity of twenty-five is evidently about midway between the greater intensities, which retard, and the smaller, which accelerate.

TABLE III.

Treatment.	Effect.
Secondary beta rays emerging from a 1 mm.-lead screen which received primary rays of an intensity of	
15	9% retardation
18	39% retardation
Averages 16½	24% retardation

Table III gives the retardations brought about by secondary beta radiations (consisting of especially slow beta electrons). The conditions were such as would have produced exposures to beta radiations of fifteen and eighteen strength had not a millimeter of lead been placed between the eggs and the radium. As a result, the beta radiations coming to the eggs were reduced in strength, i. e. there were produced secondary beta radiations, consisting of slower electrons, whose energy value was small, not only in relation to the unscreened beta radiations, but even in comparison with the smaller amount of radiations let through the lead. There was an average retardation of twenty-four per cent. The exposures of Table II, where the average intensity was greater than in these experiments, gave only slight accelerations and retardations, which balanced each other. The marked intensification of the effect by the use of the lead screen must therefore be due to the small quantity of secondary rays coming from it. The experiments therefore indicate that the secondary radiations are much more effective in proportion to their energy value than the more penetrating direct radiations.

It has been the experience of dermatologists that an unscreened X-ray tube brings about, beside the deep seated effect, a superficial burn, evidently from secondary beta radiations produced in the glass by the X-rays. A thin screen of a substance which cannot itself give rise to such secondary radiation prevents the burn by cutting off the secondary radiations arising from the glass. It is clear from these observations that the slow secondary radiations can injure tissue. A comparison of the effect of rapid and slow radiations whose relative energy values are known, has not to the writer's knowledge been previously made.

TABLE IV.

Results of treatment with secondary rays from gamma rays, produced by screening with a sheet of lead 4.5 cms. thick, 200 mgs. of 1/1000 strength of pure RBr plus 10 mgs. of pure RBr.

1	25%	acceleration
2	8%	"
3	33%	"
Average	22%	"

In Table IV a source of radiation several times as intense as in preceding trials was screened with four and a half centimeters of lead to the exclusion of all direct alpha and beta particles. Secondary beta radiations in small amounts were produced from the lead by the penetrating gamma rays which passed through it. Acceleration resulted in each of the three experiments here reported. The trial was made as a test of gamma radiation. The stimulating power of even very weak secondary radiations shown by the previous experiments indicates that the acceleration here also is, at least in part, due to secondary electrons.

SUMMARY.

The retarding effect on growth produced by beta radiations, from 100 mgs. of radium of one thousandth the strength of the pure bromide in a cubical mass placed from one to two and one half centimeters from Drosophila eggs for twenty-four hours, was more intense the nearer the eggs were to the radium. At greater distances, up to five centimeters, there was some evidence for slight acceleration.

Secondary beta radiations (slow electrons) produced a much stronger effect than primary radiations (rapid electrons) of like intensity.

Tubularia.

The exposures of Tubularia were varied not by changing the distance between the radium and the pieces of hydroid, as in the case of the Drosophila eggs, but by varying the period of exposure. Three hundred milligrams of impure radium one thousandth as strong as the pure bromide was held in a tightly closed cell (Fig. 2) consisting of a glass floor 25 mm. square, sides of sealing-wax two millimeters high, and a roof consisting of a very thin mica window. The cell rested in a Petri dish of salt water. Pieces of the stem of the hydroid six tenths of a millimeter in diameter were laid upon the window. The quality of the radiations was constant for all the experiments, since this method of exposure was always employed. The alpha rays were cut off from the hydroid stems by the mica. The gamma radiations, were so weak as to be negligible.

The pieces of stem were sixteen millimeters in length, of nearly equal diameter and from like portions of thrifty colonies. With hardly an exception they regenerated one hydranth. The pieces used as control received treatment like the others, except that the cell on which they were placed contained a non-radioactive yellow powder.

The whole process of regenerating a hydranth was arbitrarily divided into eight stages for the purpose of numerically expressing the amount

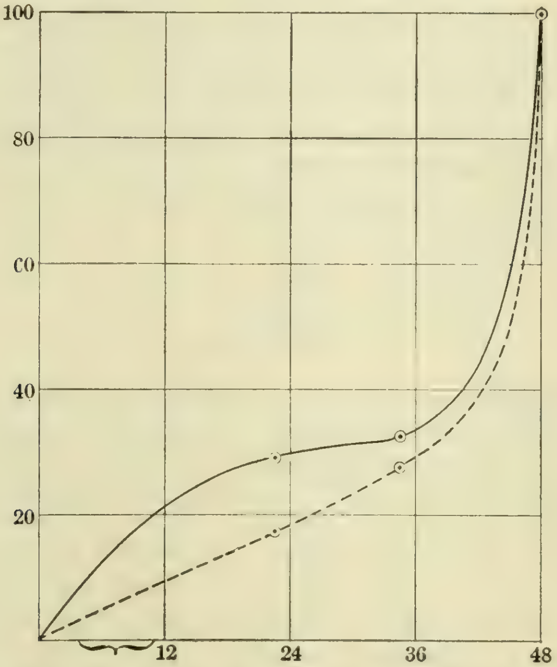

Fig. 5.— Curves showing acceleration of hydranth regeneration due to 7 hours' exposure to the beta rays of radium.

The units of the ordinate indicate per cent of development.

The units of the abscissa indicate hours from time of cutting.

The curve with the broken lines is based on control pieces.

The curve with the continuous line is based on pieces exposed to radium.

The bracket indicates the period of exposure.

of regeneration accomplished at any particular time. Thirty to fifty pieces, for the control as well as for the exposed set, were used in each experiment. At various times during the three or four days occupied in regeneration, the stage of regeneration of each piece was noted and the averages calculated for the exposed and for the control set.

The most satisfactory method of comparing the regeneration of the normal and the exposed sets was to plot the average development as a curve, using as ordinate degree of development, and as abscissa the number of hours elapsing between cutting the pieces and making the observation. When one curve was superposed on the other, the area between the curves showed the product of the degree of difference between the development of the two sets into the time during which the difference occurred (see Figs. 5 and 6).

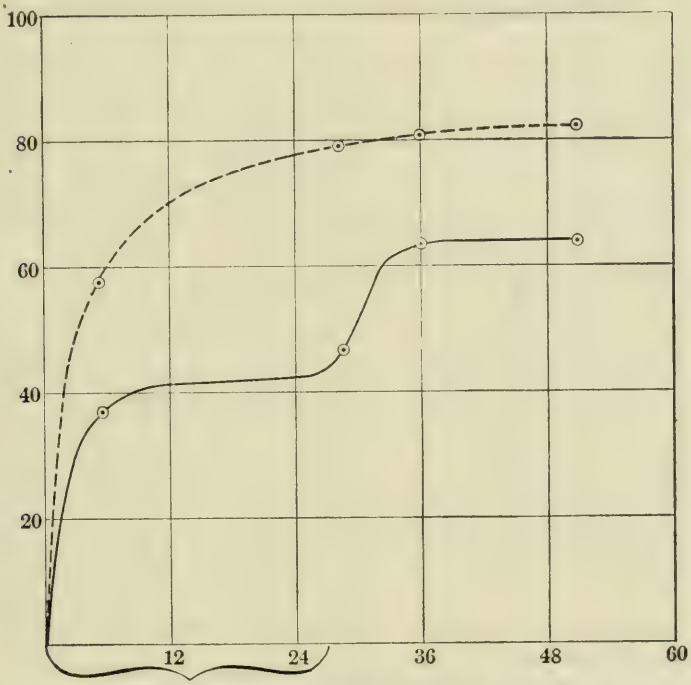

Fig. 6.— Curves showing retardation of hydranth regeneration from a 27-hour exposure to the beta rays of radium.

The units of the ordinate indicate the per cent of development.
The units of the abscissa indicate hours from time of cutting.
The curve with broken lines is based on control pieces.
The curve with the unbroken line is based on pieces exposed to radium.
The bracket indicates the period of exposure.

The area lying between the axis of abscissas and the part of the control curve extending up to the time of maximum regeneration

(Partial resorption followed regeneration) was measured and likewise the area between the two curves. The latter was expressed as a per cent of the former. The result is a quantitative expression of retardation (or acceleration) of regeneration which can be compared with similar quantities for other experiments, whereby are avoided such errors as would arise from differences between the degree of regeneration of the control in different experiments.

Acceleration and retardation were found to result from different types of exposure, as seen in Figs. 5 and 6. The conditions determining whether acceleration or retardation will occur are shown by the correlation tables (Figs. 7 and 8), which include the results of all experiments. The ordinates indicate the number of hours of exposure to radium. The abscissas in Fig. 7 stand for per cent retardation, and in the next table, for per cent acceleration calculated from the areas of the curves. The dots in Fig. 7 and the circles of Fig. 8 stand each for an experiment. The connecting of a dot to a circle is to show that acceleration and retardation occurred at different times in the same experiment.

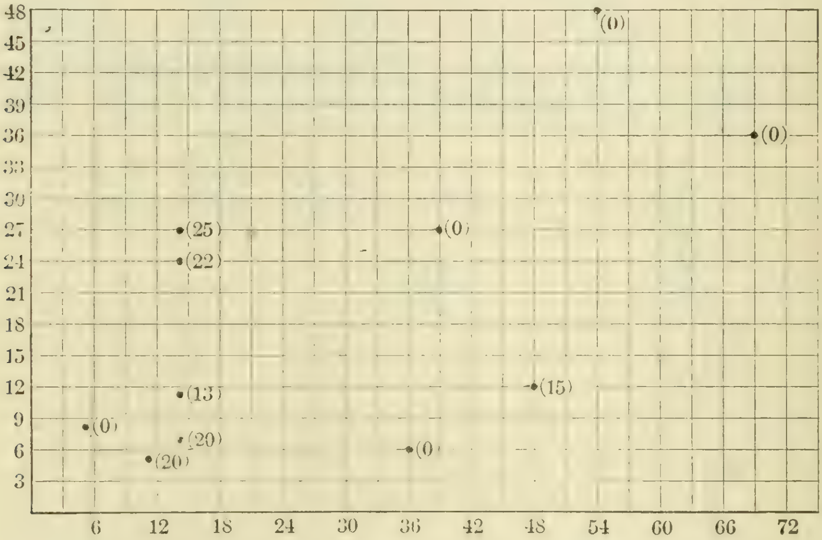

Fig. 7.— Correlation table showing the relation between length of exposure and per cent of retardation.

The units of the ordinate indicate the hours of exposure.

The units of the abscissa indicate per cent of development.

The numbers in parentheses indicate relative development before beginning of exposure.

The chief cause of the acceleration shown in Fig. 8 is apparent upon comparing the lengths of treatment in it with those which produced the retardations shown in the preceding table. Upon the whole the exposures producing acceleration were the shorter. This is an illustration of the well-known condition, shown also for growth of Drosophila eggs in the first part of this paper, that many stimuli which retard or stop growth if of high intensity will accelerate if they be weak enough. The maximum acceleration is not as great as the maximum retardation because, in the nature of things, it must be more limited.

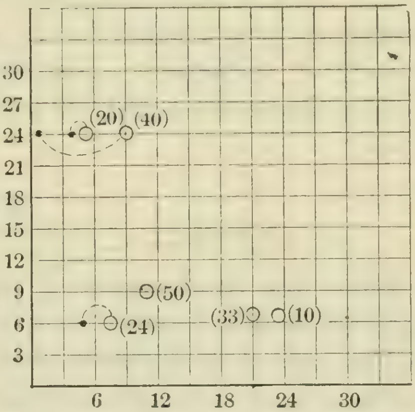

Fig. 8.— Correlation table showing the relation between length of exposure and per cent of acceleration.

The units of the ordinate indicate the hours of exposure.

The units of the abscissa indicate the per cent of development.

The numbers in parentheses indicate relative development before beginning of exposure.

Dots stand for retardation found in the course of an experiment which also showed, at a different time, the acceleration indicated by the circle connected to the dot by the broken line.

The table of retardations (Fig. 7) shows that on the whole the amount of retardation varies directly with the length of exposure; but in the acceleration table the position of the circles is too irregular to prove a correlation between these two factors.

There is much to indicate that, if time be given for the cut pieces to partially regenerate before exposure, the effect of the beta radiations is decreased. This would explain the aberrancy of the two fourteen per cent retardations from the twenty-four and twenty-seven hour exposures. It would also give significance to the fact that the five greatest retardations resulted from exposures all but one of which began at once after cutting, while for the accelerations there was an average development of twenty-nine per cent before the exposures were begun. A careful examination of the two tables makes it clear, that differing amounts of regeneration before exposure has played only a minor part in the various retardations and accelerations, yet it is true that the slope of the line of correlation in the retardation table would be less steep were a correction made for this factor. The moderate slope of the curve shows that the retarding effect does not increase so rapidly as the length of exposure, though within the limits of these exposures, it does continue to increase. In other words the sensitiveness of the hydroid decreases as the length of exposure increases. This may be simply an expression of Weber's Law, that beyond a certain maximum of intensity any stimulus has a decreasing power of stimulation. Or, it may be that the early stages of regeneration are more sensitive to exposures for the same reasons that the embryo is more sensitive than the adult.

The view is commonly met in medical writings, that the action of radium is proportional to the product of length of exposure into intensity of radiation. The Figure 7 correlation table shows clearly that, as far as the growth of hydroids is concerned, that view is erroneous.

Summary.

When the fundaments of regenerating Tubularia hydranths were exposed to beta radiations from three hundred milligrams of impure radium one thousandth as strong as the pure bromide for periods up to three days in length, the shorter exposures were found to accelerate regeneration and the longer to retard. The degree of retardation increased slowly with lengthening exposure; but the degree of retardation relative to the length of exposure decreased with lengthening exposure.

Bulletin of the Museum of Comparative Zoölogy
AT HARVARD COLLEGE.
Vol. LIII. No. 8.

EFFECTS OF RADIUM ON LIVING SUBSTANCE.

II. COMPARISON OF THE SENSITIVENESS OF DIFFERENT TISSUES IN THE DUNG-WORM ALLOLOBOPHORA FOETIDA, AND IN THE CRAYFISH CAMBARUS AFFINIS, TO THE BETA RAYS OF RADIUM.

By E. D. Congdon.

CAMBRIDGE, MASS., U. S. A.:
PRINTED FOR THE MUSEUM.
February, 1912.

No. 8.— CONTRIBUTIONS FROM THE ZOÖLOGICAL LABORATORY OF THE MUSEUM OF COMPARATIVE ZOÖLOGY AT HARVARD COLLEGE, UNDER THE DIRECTION OF E. L. MARK, No. 229.

Effects of radium on living substance.— II. *Comparison of the sensitiveness of different tissues in the dung-worm Allolobophora foetida and in the crayfish Cambarus affinis to the beta rays of radium.*

By E. D. Congdon.

The relative sensitiveness of different tissues of vertebrate animals to beta radiations has been determined with a fair degree of exactness (see Thies, :**05**, Lossen, :**07**), though contradictory results have been obtained for some tissues, especially for the central nervous system. The worm Allolobophora foetida was taken as one of the objects of exposure and the histological changes examined in twenty-four individuals to see whether there was agreement in the relative sensitiveness of their different tissues and the corresponding tissues of vertebrates. As the largest animals were only three millimeters in diameter and the whole length of the body was laid in the grooved cover of the radium cell, all parts of the body, and thus all tissues, received a nearly equal amount of radiations. The debated question of the sensitiveness of the central nervous system was therefore put to the test under especially favorable conditions with Allolobophora, as it also was with Cambarus.

Allolobophora foetida.

For the exposure of Allolobophora a cell (Figure) was used three millimeters in depth, two centimeters wide, and four centimeters long, containing three hundred milligrams of impure radium of one thousandth the strength of the pure bromide.[1] The roof of the cell was a very thin sheet (window) of aluminum, the upper surface of which was coated with a delicate film of paraffin. This sheet was bent so as to form a longitudinal trough, in which the animal was laid surrounded by a piece of paraffined brass netting bent into the form of a tube. Wet filter paper was laid over the netting to keep the worm moist. The occasional change of position on the part of the worms during treatment and the production of secondary radiations from the netting above them both contributed to still greater uniformity of exposure. The gamma rays were so weak as to be negligible. For each experiment two individuals of equal size were selected, one for exposure to radium, the other for the control experiment. The one not irradiated was placed over an empty cell and in every way, except for exposure to radium, treated like the irradiated animal. The two worms were then fixed and stained at the same time and in the same manner.

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[1] For the use of a part of this radium I am under deep obligation to Mr. Hugo Lieber, who placed it at the disposal of Professor Mark for the purpose of aiding in these investigations. To Dr. Theodore Lyman I wish to express my appreciation for his kindness, not only in loaning radium, but also for advice in matters of radium physics.

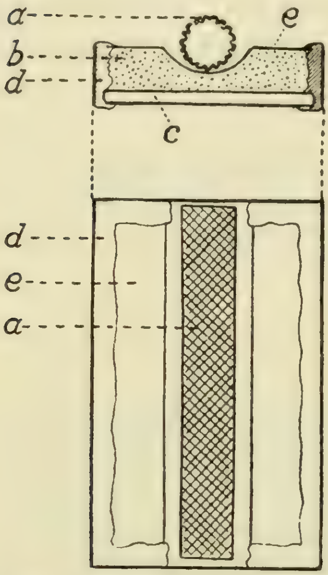

Fig. 1.— Diagram of cell containing the radium and the cylindrical wire cage to hold the worm used in experiments on Allolobophora. Above, the apparatus in cross section; below, plan. *a*, Wire cage; *b*, radium; *c*, glass floor of cell; *d*, sealing-wax rim; *e*, aluminum roof of cell, so bent as to form a trough to receive the caged worm. Magnified two diameters.

Individuals in three different stages of development were selected for treatment: (1) immature worms of approximately one tenth the bulk of the smaller adults; (2) adults of rather small size, found to have a well developed clitellum and reproductive organs; (3) large worms with reproductive organs active, taken while copulating.

That radium has an effect upon Allolobophora is shown in the accompanying table by the death of ten out of twenty-four of the

TABLE.

Number of experiment.	Strength of radium.	Hours on radium	Duration of life after radium.	Fate.
		Young Worms.		
1	Weak	3½	0 hrs.	Dead
2	"	5½	3 "	"
3	"	1½	4½ days	Killed
4	"	6½	2¼ "	"

Number of experiment.	Strength of radium.	Hours on radium	Duration of life after radium.	Fate.
		Small Adult Worms.		
5	Weak	3¼	0 days	Killed
6	"	5	0 "	"
7	"	4½	96 "	"
8	"	6	5 "	"
9	"	6	2 "	Dead
10	"	7	0 "	"
11	"	6	30 "	Killed
12	"	9½	0 "	"
13	"	12	4 "	"
14	"	15	30 "	"
15	"	15	33 "	Dead
16	"	15	48 "	"
		Copulating Worms.		
17	Weak	12	0 days	Dead
18	"	4	120 "	Killed
19	Strong	3	144 "	Dead
20	"	48	120 "	Killed
21	"	20	96 "	"
22	"	26	72 "	Dead
23	"	24	168 "	"
24	"	24	120 "	Killed

animals which were exposed. Most of the other fourteen gave signs of having suffered from the exposure before they were killed. Some became insensitive to tactile stimulus. Others showed alternate swellings and contractions along the body. One animal turned markedly browner than any of the others, although otherwise it looked normal. There was some evidence of a darkening of other exposed animals, which was confirmed by a microscopic study of the body wall. The effort was made to fix the worms after degeneration was apparent in their tissues, but before death. That this was difficult to do, is indicated by the fact that half of the animals killed showed no internal injury, although they did show already some of the external indications which have been mentioned. A comparison of the periods necessary to produce death of the animals of different ages, as represented in the three subdivisions of the table, shows that the young animals succumb much more quickly than the adult. This may be due either to the greater sensitiveness of the tissues of the young, or to a simple physical condition, namely that, since the diameter of the bodies of the young were less than half that of the old, they must have absorbed more radiation in proportion to their volume than the adult. The irregularities in the effects produced by like exposures must be due to differences in the animals, since the character of the exposure was unvarying.

The degenerative changes found at the three stages are similar, and so may be described together. An excess of pigment was observed in the body wall in a number of cases. One small adult at the end of four and a half days had turned a reddish brown, markedly darker than the color of any other animal; several other worms showed coloring of less intensity. The pigment which produced the coloring occurs normally in the body wall, but in small amounts. A microscopic examination shows it among the connective-tissue cells in which the circular muscle cells are imbedded. Inasmuch as blood sinuses appear in the same regions when the body wall happens to be gorged with blood, and pigmentation is often produced by blood, it seems probable that the pigment in these spaces is haematogenic.

The testes of copulating worms were strongly affected by the rays. There was an entire loss of spermatogonia and a scarcity of the first spermatocyte stage. Their places were occupied by vacuolated spermatophore cells, whose nuclei showed chromatolysis. Some nuclei of the primary spermatocytes, and possibly of spermatogonia, were represented by swollen or shrivelled nuclear walls containing no chromatin. I did not find the degeneration in its early stages. The elongating secondary spermatocytes and the contents of the seminal vesicles appeared normal. In parts of the testis a condition of their further degeneration was found in which some chromatin had gone into solution. In the testis of a copulating worm which had died as the result of radium treatment less than two hours before it was put into a fixing fluid, most regions of the germinal tissue were a mass of cell debris devoid of chromatin. Here and there the spermatocytes of very resistant tubules were indicated by rounded cells whose nuclei stained deeply in haematoxylin. Spheres unstained by haematoxylin were present and were taken to be the archoplasmic masses. The other tissues of the animal showed no evidence of degeneration previous to death.

It has been found that the beta radiations of radium, as well as X-rays, are especially destructive to the spermatocytes of the mammalian testis (Thaler, :**05**; Thies, :**05**; London, :**05**; Albers-Schönberg, :**03**; and others). In the earthworm testis, spermatogonia and spermatophore cells had degenerated, as well as the spermatocytes.

No sufficiently early condition of degeneration was found to determine whether the spermatocytes are the first to suffer, as in the mammalian testis.

The ovaries of several worms were in a necrotic condition. The usual effect was a clumping and dissolving of the chromatin of the oögonia. The oöcytes were more resistant. The most advanced stage of degeneration was accompanied by a shrinkage of oöcytes, or a loss of their boundaries by a granular degeneration. Necrosis of the mammalian ovarian follicles due to beta radiations has been described by Halberstaedter, **:05**; Bergonié, Tribondeau et Récamier, **:05**; London, **:05**; and others. They find that the oöcyte-stage, like the corresponding stage in the testis, is the most sensitive. It is noteworthy therefore that in the earthworm the oögonia were more affected.

Destruction of the digestive epithelium was common and especially noticeable in the young worms. In the epithelial cells of the anterior part of the digestive tract of the adult there was a loss of cell boundaries, a vacuolation, and nuclear degeneration. The basal cells were at times similarly affected, or their substance was broken down into granules. The abnormal condition of the digestive epithelium was associated with a distension of the lumen of the intestine by a liquid. The radium may have acted by causing an unusual osmotic condition which resulted in filling the stomach-intestine; or the digestive secretions may have been weakened so that bacteria were able to thrive and produce putrefaction.

In mammals the digestive tract as a whole has been usually considered a region little sensitive to radium. Perhaps this opinion has arisen partly because of the protection from the radiations afforded by its central position in the body. In Allolobophora, on the contrary, the small diameter of the body results in an action of the radiations on the stomach-intestine almost as intense as that on the body wall. It is not possible to say that the degeneration of the digestive epithelium is entirely due to the direct action of the radiations upon its cells, since pressure of the fluid distending the lumen and the chemical action of the fluid may have hastened the destruction.

The body wall was often broken down on one side of the animal, presumably that which was longest near the radium. The lesions were found in regions where the genital glands or intestine were affected, but as they also occurred elsewhere, they were not necessarily a result of the degeneration of those organs. The muscle plates of muscle cells became massed together and the chromatin of both connective-tissue cells and muscle cells was dissolved. The epidermal cells were vacuolated and their nuclei were undergoing chromatolysis. The gland cells had often lost their contents.

Especial attention was given to the condition of the supra-oesophageal ganglia and nerve cord, because of the differences of opinion as to the sensitiveness of the central nervous system to radium.

Danysz (:03) exposed the brain of the mouse to radium after trepanning the skull, and he described as a result the necrosis of the nerve cells. Rehns (:05), upon the other hand, found little injury in the brain of a rabbit with skull trepanned. Bohn (:03), Scholtz (:04), Obersteiner (:04), and others describe an especially marked degeneration in the central nervous system of animals whose entire bodies had been exposed to radium. Birch-Hirschfeld (:05) found it to be the nervous elements of the rabbit's retina which most quickly succumb to beta rays. The fact that the central nervous system readily degenerates does not, however, prove that it is especially sensitive, since it contains much vascular tissue and this is well known to be highly sensitive to radium. Observers have been divided as to whether the degeneration of nervous tissue was actually a primary effect of the radiation, or produced by the necrosis of the vascular endothelium and the resulting hemorrhage.

In the earthworm the nerve cord is associated with much less vascular tissue than is the spinal cord in the mammal. This makes it possible to ascertain whether the nervous tissue is of itself especially sensitive to radium. A careful examination showed that the ganglia and nerve cord are no more sensitive than the body wall, and were necrotic only when it also was affected. The ganglion cells were, naturally, the first elements of the nerve cord to show injury. Later the fibre tract lost its structure and the nuclei of the neuroglia cells were broken down. The Nissl flakes did not give evidence of any injury to the nerve cord or ganglia in cases where the body wall remained normal. Furthermore, pigment deposits were found in the body wall of worms whose nerve cord was in good condition. Inasmuch as the pigment is probably a product of decomposition of the blood, this is an indication that the vascular system, as in mammals, is very sensitive to beta radiations.

The Nerve Cord of the Crayfish Cambarus affinis.

In the crayfish (Cambarus), as well as in Allolobophora, the central nervous system is much freer from vascular tissue than in mammals, and furnishes, therefore, a favorable object for determining whether the nerve cells themselves degenerate quickly when free from necrosis of the blood vessels and from hemorrhage.

It was found possible in Cambarus to bring a small radium cell, without a groove, but otherwise similar to that used for Allolobophora, into direct contact with the ganglia of the ventral nerve cord. In this way the intensity of the beta rays which fell upon the ganglia was known, because there was no absorption of them by intervening tissues. In order to prevent the cord from being injured by possible earlier degeneration of the tissues surrounding it, the treatment was made short but intense.

In each experiment one to three ganglia of the ventral nerve cord of two animals were exposed by removing some of the abdominal sclerites. The opening was made just large enough to accommodate a small radium cell containing thirty milligrams, of radium one thousandth as strong as pure radium bromide. A dummy cell of the same size and shape was used for the control animal. It was found possible to apply the cell to the cord with little bleeding. The animals were kept motionless by tying abdomen and caphalothorax to a board. A slight current of water was allowed to pass over them to permit respiration. Especial care was taken to prevent pressure or other mechanical disturbance to the cord. Immediately upon the conclusion of an exposure, the condition of the tissues about the cord was noted. The ganglia were then immediately removed and fixed in 95 % alcohol, after which they were stained in toluidin blue and erythrosin to differentiate the Nissl flakes.

Pure radium bromide was applied in one experiment for eighteen hours. Four animals were exposed in succession, each for twenty-four hours, to the cell of weak radium and another to ten milligrams of very pure bromide contained in a glass tube. There was no necrosis produced by the rays in either ganglion cells or fibre tracts. The Nissl flakes, because of their promptness in responding to various forms of nervous injury, were examined carefully for peculiarities of any kind. They did not differ from those in unexposed ganglia of animals treated in a similar way with a dummy cell.

As it is the common experience of dermatologists that exposures to intense beta rays produce degenerative effects in a very short time, and since the Nissl flakes show injury so quickly, it is not likely that the ganglion cells in these experiments were fixed during a latent period precedent on degenerative change. The same cell of weak radium used for these exposures produced a marked effect upon frog embryos in the yolk-plug stage within forty-eight hours after exposure.

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Bulletin of the Museum of Comparative Zoölogy
AT HARVARD COLLEGE.
Vol. LIII. No. 9.

ON THE HAIR-LIKE APPENDAGES IN THE FROG ASTYLOSTERNUS ROBUSTUS (BLGR.).

By Willy Kükenthal.

With Five Plates.

CAMBRIDGE, MASS., U. S. A.:
PRINTED FOR THE MUSEUM.
February, 1912.

No. 9.—CONTRIBUTIONS FROM THE ZOÖLOGICAL LABORATORY OF THE MUSEUM OF COMPARATIVE ZOÖLOGY AT HARVARD COLLEGE. E. L. MARK, *Director*, No. 230.

On the hair-like appendages in the frog Astylosternus robustus (Blgr.).

BY WILLY KÜKENTHAL.

While working at the Museum of Comparative Zoölogy at Harvard University, soon after my arrival in Cambridge, Dr. Thomas Barbour called my attention to some frogs collected in Kamerun. I was much struck by their very peculiar appearance which showed a dense covering of hair-like filaments on both sides of the body and on the outer surface of the thigh (Plates **1**–**3**).

This frog was described by Boulenger (:**00**, p. 443, pl. XXX), who named it Trichobatrachus robustus, thus establishing, not only a new species, but also a new genus. Boulenger pointed out that the villose dermal papillae covering some parts of the body are far from being a nuptial attribute of the males, as one might have been inclined to suppose from analogy with various fishes, and he emphasized the fact that this character is more strongly developed in the female than in the male. He suspected, therefore, that these hair-like appendages are a mere seasonal peculiarity. About a year later Boulenger (:**02**) published another short paper upon the same subject, having meantime had the opportunity of investigating seven more specimens besides the two he had at first. He refers to an examination of the histological structure of the hair-like appendages made by Dr. H. Gadow (:**00**) and confirms Gadow's view that we possess no clue whatever to their physiological signification. Five out of the seven specimens were adults. Of these five adults, three were females. Although evidently obtained during the breeding season, these females showed no trace of the appendages, whilst the two males had them very well developed.

We see that this statement is quite contradictory to that which Boulenger had made in his first publication, but, strangely, the author gives no explanation of this discrepancy.

In Gadow's short communication about the histological structure of these peculiar villosities, he tells us that males and females possess on their flanks, as well as on the upper and hind surfaces of the thighs, numerous finger-shaped, membranous, dark-colored prolongations of the skin, with a dense connective-tissue axis. In this part of the cutis he found some small insignificant blood vessels and also lymph-spaces, but neither nerves nor nerve-terminations. Therefore, in his opinion, these appendages are not to be considered as a sensory apparatus. Their function is unknown, and it also is unknown whether they are developed exclusively during the breeding season.

That is all that we know about these remarkable structures, and I have only to add, that recently Nieden (:07, and :08, p. 659) has pointed out that the genus Trichobatrachus is identical with the genus Astylosternus, the latter name having been previously (1898) employed by F. Werner.

No less than eleven specimens of this genus have been at my disposal, all coming from Kribe, Kamerun, and all belonging to the same species, Astylosternus robustus (Blgr.).

From the study of these specimens I have established: First, that the hair-like appendages are present only in the males; they are wanting in all females. That agrees exactly with the second statement of Boulenger, but disagrees entirely with his first statement, that the character is more strongly developed in the female than in the male. As he based this strange assertion on the investigation of only two specimens, a male and a female, I feel sure that some mistake must have happened. I conclude therefore, that these appendages occur only in the males, and that they are certainly secondary sexual characters.

Secondly, that these appendages do not attain the same degree of development in all male individuals, and that even in full grown males there are very conspicuous differences in this regard.

In one jar there were preserved three adult males, apparently captured at the same time the largest being 115 mm. long, and all these large males showed very short appendages, in some places merely suggestions of appendages (Plate **4**). Another jar contained two adult males of about the same size as the three, one specimen being, however, somewhat smaller than the three contained in the first jar, since it measured only about 100 mm. in length. The two larger specimens had a very remarkable appearance, caused by their very long hair-like villosities, which attained in the larger of the two (Plates **1–3**) a length of about 20 mm. In the same jar was also preserved a third, much smaller, male, measuring only 80 mm.; but this also was provided with short, though quite distinctly developed appendages.

Thus we see that in this species there are males with long and males of the same size with very short appendages. I find no males without these villosities, for even the smallest male at my disposal possesses them.

It is to be regretted that the collector of these specimens, Mr. Schwab, has not given any notes about the time of year when he captured the different specimens; but, notwithstanding this, we can draw with almost absolute certainty the conclusion that these appendages are much more highly developed at one time of year than at other times. Moreover it is very probable that this time corresponds with the breeding period. Direct observations, of course, would quickly settle this question.

The fact that a younger (smaller) male, contained in the same jar with the two adult males possessing fully developed appendages — and therefore apparently captured at the same time with these — showed this hairy coat in its beginnings, points to the conclusion that the appendages are fully developed only on adult animals, and probably, as I have already suggested, at the time of mating.

Now arises the question, from what do these organs originate? The reply requires a careful investigation of the female (Plate **5**). It is quite surprising, that none of the former investigators has observed the fact, that the females have, on exactly the same parts of the body that on the males bear these appendages, small but quite distinct tubercles, which have the same diameter as the bases of the appendages in the male. Their distribution over exactly the same areas of the surface shows clearly that they are homologous with the appendages of the males.

Moreover, if we carefully study the surface of the skin (Plates **1**, **3**–**5**), we find that both males and females show similar tubercles scattered over the whole back, and that they are more closely crowded in the region of the angle of the jaws (Plate **3**). In some areas of the surface of males we may even observe the transition of these tubercles into the villous appendages. From these comparisons we must therefore draw the conclusion that these appendages have originated from tubercles of the skin, such as we find scattered over the skin of this species in other regions of the body and such as are recorded from other species of Ranidae.

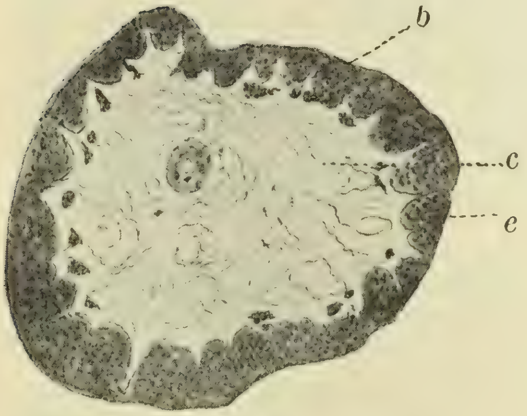
These hair-like appendages are therefore to be considered as highly developed tubercles of the skin.

I have studied the microscopical structure of these appendages in series of sections, which Mr. S. Kornhauser has been kind enough to prepare. The stains which were used were: borax-carmine and bleu de

Lyon, Ehrlich's stain and picric acid-fuchsin, and Ehrlich's stain and Orcein; impregnation after Bielschowsky was also used.

The results at which I have arrived after studying these microscopical preparations are entirely different from those which Gadow published in his paper already referred to. Each appendage consists of an inner cutis papilla and an epidermal outer layer of very peculiar appearance. This outer layer is made up of many longitudinal ridges of epidermis cells, between which are found deep longitudinal grooves. These grooves are filled with the cutis tissue. In transverse section (Fig. A) this condition is very conspicuous, reminding one somewhat of the transverse section of a developing feather.

The stratum corneum is not thick, but quite distinct, and outside this is still another continuous external layer of horny cells, which form a kind of loose covering, thus indicating that these appendages, like the rest of the skin, periodically slough their superficial stratum corneum. This statement does not agree with Gadow's sentence: "Die Epidermis ist weich und weder besonders verdickt noch irgend wie verhornt." On the other hand, I was not able to find the great number of glands which Gadow describes; at least, they are not more numerous than in other parts of the skin.



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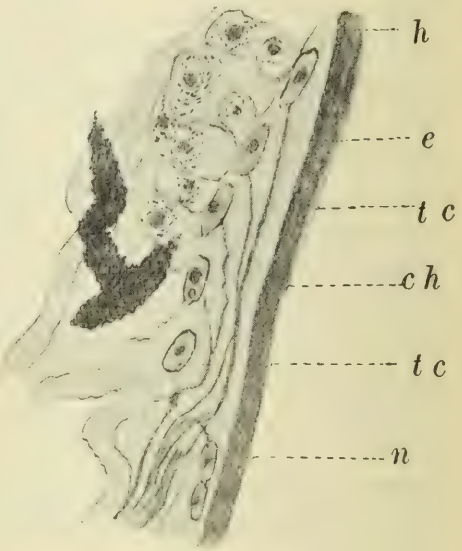

B

Fig. A. Transverse section of one of the hair-like appendages. *b*, blood vessel; *c*, cutis; *e*, epidermis.

Fig. B. Portion of a longitudinal (radial) section of hair-like appendage, showing tactile cells in one of the cutis ridges. *ch*. chromatophore; *e*, epidermis cells; *h*, stratum corneum; *n*. nerve fibre; *tc*. tactile cells.

There is a quite conspicuous blood vessel running along the axis of the cutis papilla and other smaller blood vessels are found in the surrounding substance of the cutis. The whole papilla is built up of a dense connective tissue, whose fibres run for the most part either in transverse or in longitudinal directions. Chromatophores are numerous, being especially abundant at the base of the appendage.

Dr. Gadow denies that there are nerves or nerve-terminations in these appendages and therefore maintains emphatically that the function of sensory-organs is wholly excluded from these appendages. In opposition to that, my own observations have convinced me, that there are both nerves and nerve-terminations in these appendages, and that therefore they do serve as sensory organs.

Owing to the fact that the objects were preserved in alcohol, the impregnation by Bielschowsky's method did not work well; but I finally succeeded in observing large nerves (Fig. B) entering the papilla from its base, and in finding some tactile cells (*tc.*) of the same shape as those described by Merkel in other frogs. They were situated in the grooves between the epidermal ridges. Here the epidermis is very thin. The tactile cells appeared as rather flat protoplasmatic bodies with a quite distinct nucleus, and their broader sides were parallel to the outer surface of the epidermis. Each of these tactile cells was provided with an axis-cylinder, which ran quite close to the surface of the epidermis, but in the cutis tissue beneath it, and were united proximally into a common nerve fibre (*n*). The nerves of these tactile cells therefore come from the outer, not from the deep, surface of the cells, and this agrees very well with some statements made by Merkel (**'80**, p. 109) regarding the innervation of tactile cells. The condition of my material did not allow me to carry this investigation further, but from the evidence I have, I maintain that these appendages contain nerves and nerve terminations and, therefore, that they must have something to do with sensory functions. Besides these, there may be other functions, it is true, as is suggested by the presence of glands.

It is also a striking fact that these appendages appear only on those areas of the surface where, according to Merkel (**'80**, p. 108, Taf. IX, Fig. 2), in other frogs these tactile cells ("Tastflecke") form aggregations.

The results, then, of this investigation are, that these appendages in Astylosternus robustus appear only in the males during the mating season, and are to be considered as secondary sexual organs of a very peculiar hair-like shape, originating from tubercles of the skin, and that they are charged with sensory functions.

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EXPLANATION OF PLATES.

PLATE 1.

Full grown male specimen of *Astylosternus robustus* (Blgr.), dorsal view.

PLATE 2.

The same specimen, seen from the side.

PLATE 3.

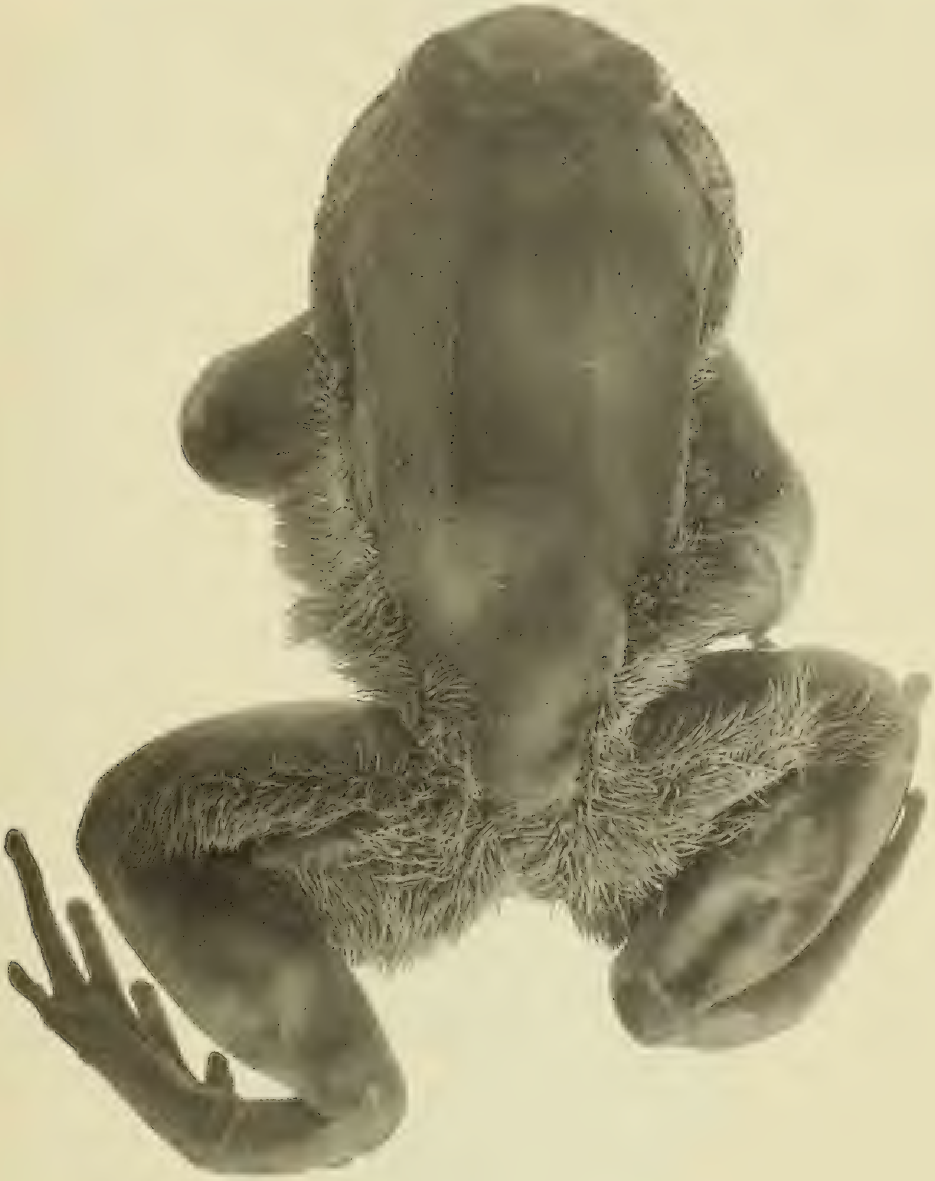
The same, ventral view.

PLATE 4.

Full grown male specimen with very small appendages.

PLATE 5.

Female specimen, showing scattered tubercles on the back.



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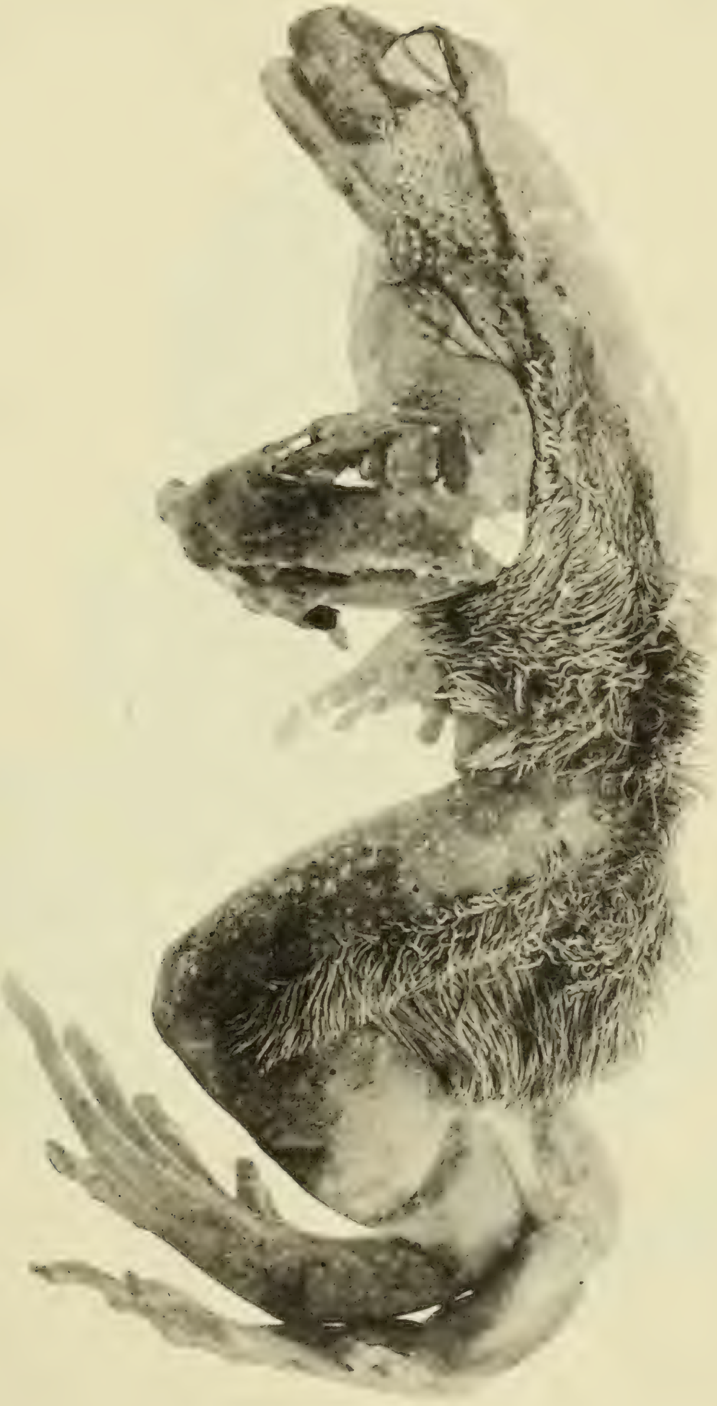

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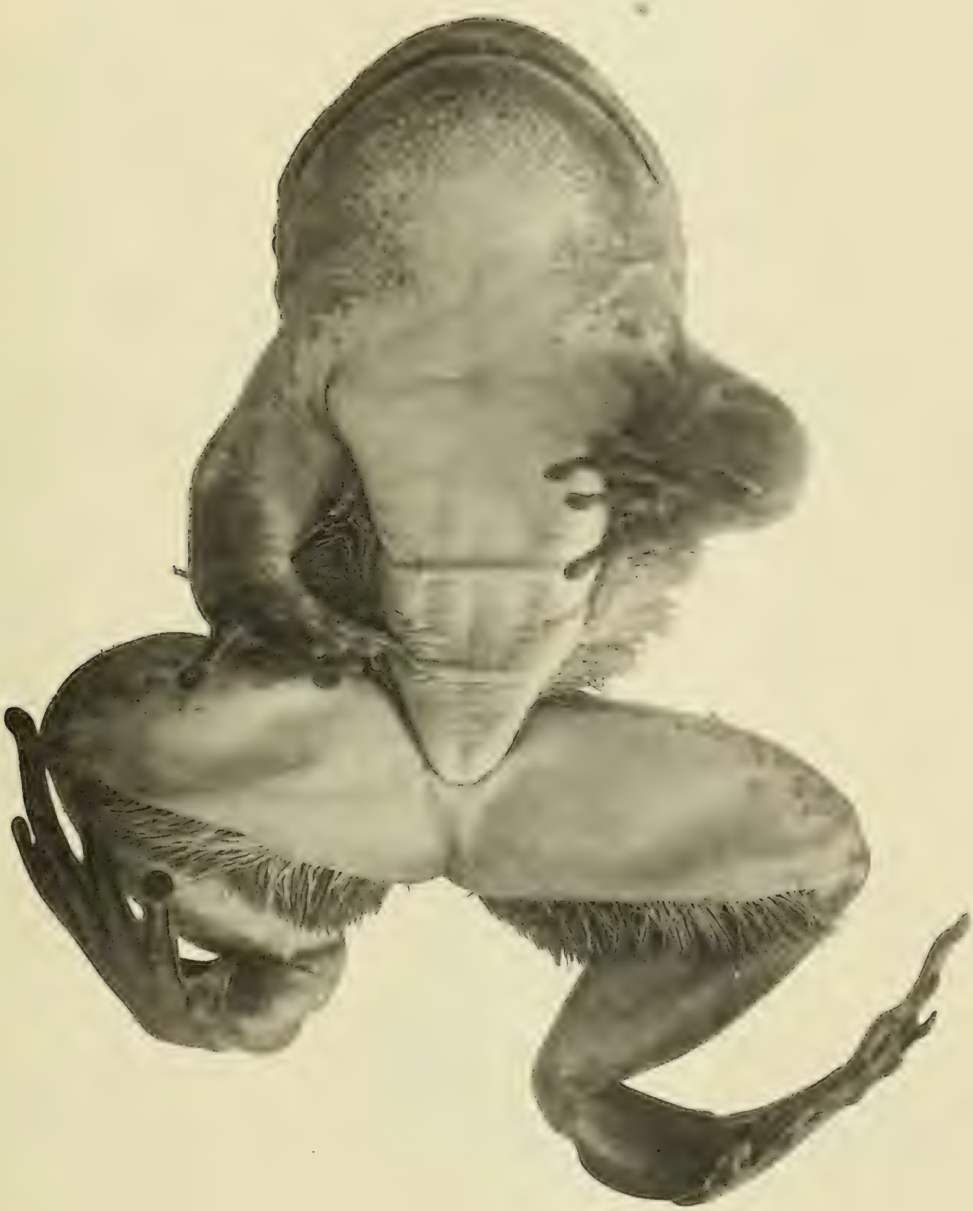

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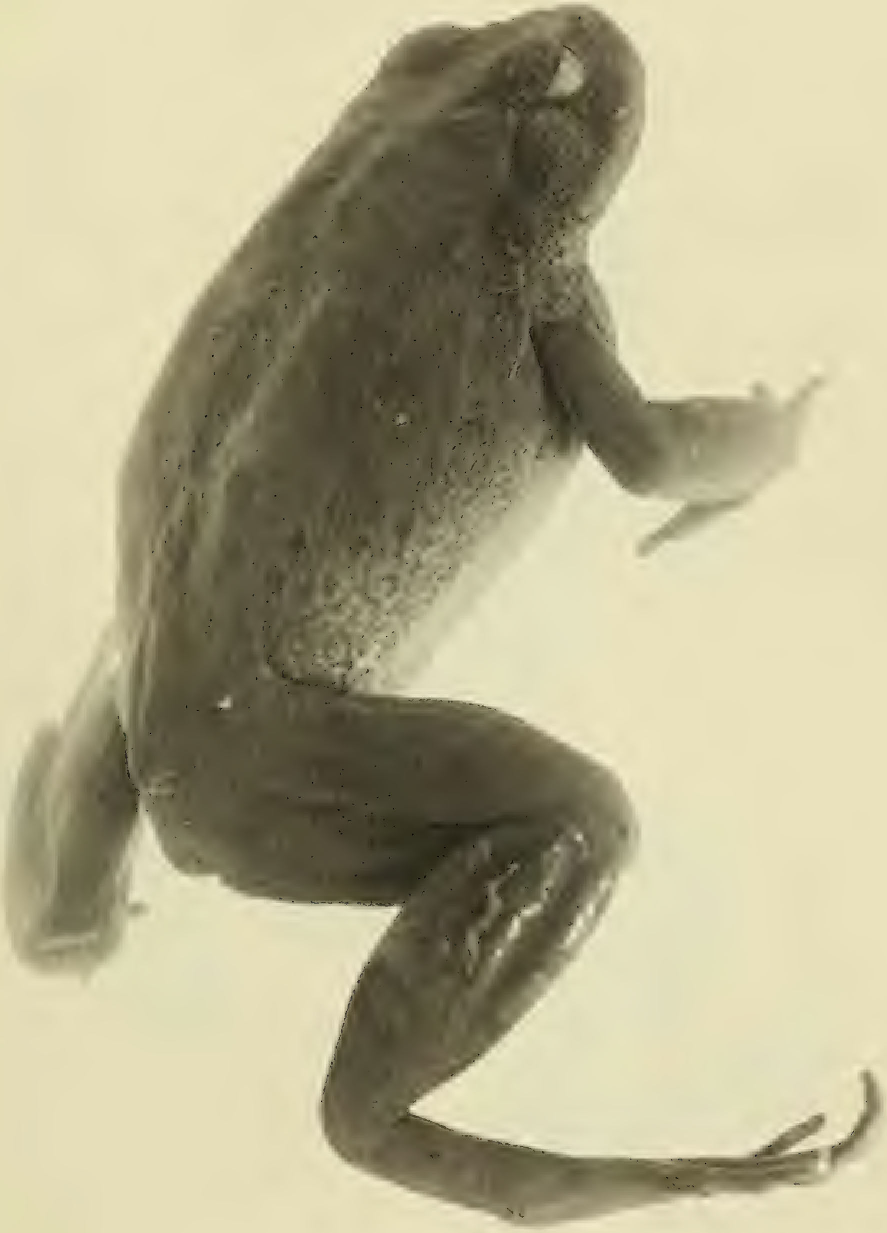

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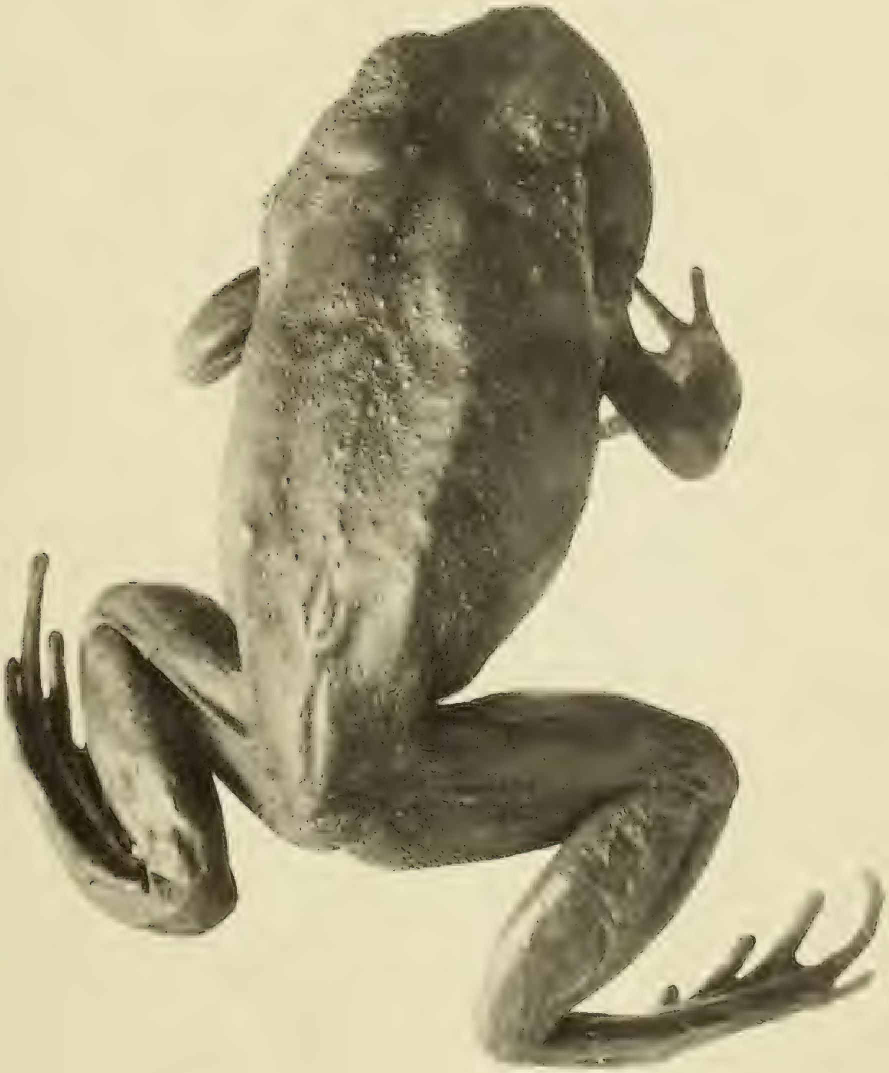

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Bulletin of the Museum of Comparative Zoölogy
AT HARVARD COLLEGE.
VOL. LIII. No. 10.

A REVISION OF THE ANTS OF THE GENUS FORMICA (LINNÉ) MAYR.

BY WILLIAM MORTON WHEELER.

CAMBRIDGE, MASS., U. S. A.,
PRINTED FOR THE MUSEUM.
OCTOBER, 1913.

No. 10.— *A Revision of the Ants of the Genus Formica (Linné) Mayr.*[1]

By William Morton Wheeler.

No revision of the American ants of the circumpolar genus Formica has been published for many years, notwithstanding the fact that it comprises some of the most important members of our insect fauna. Mayr,[2] who in 1886 first attempted a revision of this genus, cited only seven species and seven varieties from North America. We are indebted to Emery, however, for the first really serious account of these ants. In 1893 this investigator gave us, in a very succinct and admirable paper,[3] a critical account of all the known American forms, on the basis of collections made by Mr. Theodore Pergande and Rev. P. J. Schmitt. In this paper eight species, twelve subspecies, and fifteen varieties are recognized as being peculiar to our fauna. During the twenty years that have since elapsed a much greater amount of material has found its way into public and private collections, and during the past thirteen years I have described several species and have accumulated both through my own efforts and through the very generous aid of many correspondents so large a collection of Formicae, that it seems advisable again to "take account of stock" of the North American forms. A study of all this material enables me to recognize thirty-one species, nineteen subspecies, and forty-three varieties as belonging to our fauna. Since many of these are very closely related to the Palaearctic or Eurasian forms I have included a brief account of the latter in the present paper. In this part of my work I have made extensive use of Emery's recent revision of the Palaearctic ants.[4]

The species of Formica can be readily distinguished from the species of the other genera of the subfamily Camponotinae by the following characters:—

The workers are small or medium-sized ants, often varying considerably in stature but only feebly polymorphic. Their mandibles have a broad, dentate apical border. The maxillary palpi are 6-jointed, very rarely 5-jointed, the fourth joint not longer or but slightly longer than the fifth; the labial palpi are 4-jointed. The clypeus is trapezoidal and usually distinctly carinate; the clypeal and antennal foveae are confluent. The frontal area is usually very distinct; the frontal carinae are subparallel or diverging behind. The eyes are convex, moderately large, and situated behind the median transverse axis of the head. The ocelli are always distinct. The antennae are 12-jointed and inserted near the posterior corners of the clypeus; their funiculi are more or less thickened apically but without a club. The thorax is distinctly and often deeply constricted in the mesoëpinotal region. The epinotum is angular or rounded in profile and always unarmed. The petiole is scale-like, erect and compressed anteroposteriorly.

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[1] Contributions from the Entomological Laboratory of the Bussey Institution, Harvard University. No. 59.

[2] Die formiciden der Vereinigten Staaten von Nordamerika. Verh. Zool. bot. ver. Wien, 1886, **36**, p. 419–464.

[3] Beiträge zur kenntniss der nordamerikanischen ameisenfauna. Zool. jahrb. Syst., 1893, **7**, p. 633–682, 1 pl.; 1895, **8**, p. 257–360, pl. 8.

[4] Beiträge zur monographie der formiciden des paläarktischen faunengebietes. (Hym.) Teil. 7. Deutsch. ent. zeitschr., 1909, p. 179–204, 16 figs.

The female is usually considerably larger than the worker, but in some parasitic species, of the same size or even smaller than the largest worker forms. The anterior wings have a discoidal and a single closed cubital cell.

The male is always larger than the worker and usually slightly smaller than the female. The mandibles are narrow, flat, and pointed, with short, dentate or edentate apical border. The frontal carinae are very short or vestigial; the antennal scapes long, the first funicular joint longer than the second. The petiole is thicker and less compressed anteroposteriorly than in the worker and female. The genitalia are robust and conspicuous, their stipes simple, without an appendage; the subgenital plate is simple or feebly lobed. The cerci are well developed.

Ruzsky[1] was the first to divide the genus Formica into subgenera by basing a subgenus, Proformica, on the Palaearctic *F. nasuta* Nylander. He also included *F. aberrans* in the same group. All the other species he referred to the subgenus Formica *sens. str.* More recently several additional Palaearctic species of Proformica have been brought to light by Emery and Forel. As now defined, the group is based mainly on the greater length of the first funicular joint of the worker and female and of the genital stipes of the male. But the group is, on the whole, rather vague, for the recently discovered Tunisian *Proformica emmae* Forel has close affinities with Cataglyphis (Myrmecocystus *olim*), and our North American *F. neogagates*, which has several of the characters of Proformica, is in general habitus more like a true Formica. Emery [1] and I, however, have independently reached the conclusion that this ant is properly a Proformica.

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[1] The ant fauna of the Kirghiz Stepps. (In Russian). Horae Soc. ent. Ross., 1903, **36**, p. 294–316).

A study of our North American Formicae shows that *F. pallidefulva sens lat.* is even more worthy than Proformica of ranking as a distinct subgenus, for the male differs from that of the other species in much the same manner as does the male of Proformica, while the worker in the structure of the thorax and antennae is even further removed from the species of the subgenus Formica. I have therefore erected a new subgenus, Neoformica, to include *F. pallidefulva* and its various subspecies and varieties and *F. moki* Wheeler. The latter form is provisionally placed in this group because its male and female phases are still unknown.[2]

It is possible, as Emery clearly showed, to separate the various species of the subgenus Formica into groups. These are more sharply defined in the present paper. The *rufa*-like species with diminutive females I regard as constituting a distinct group (*microgyna* group), although I am unable to find any satisfactory worker characters on which to base it. The *exsecta* group is so sharply defined as scarcely to admit of discussion. I have expanded the *sanguinea* group by including in it a number of species with notched clypeus though lacking the parasitic or slave-making habits of the typical *sanguinea*. These species may have to be placed in a group by themselves when our knowledge of their sexual phases is more advanced. The *rufa* group, especially in North America, presents the greatest difficulties in the delimitation of species. This was clearly recognized by Emery, who would be the first to admit that his treatment of our *rufa* forms was inadequate on account of the insufficient amount of material at his disposal. I have endeavored to reduce the confusion by recognizing the Eurasian *truncicola* as a distinct species and by referring to it a number of forms (*integroides, integra,* and *obscuriventris*) which have been hitherto regarded as subspecies or varieties of *rufa sens. str.* The habits of all these American forms agree very closely with those of the Eurasian *truncicola* and differ from those of *rufa* and its subspecies *pratensis.* In order to simplify the treatment of the forms in the *fusca* group I have proceeded in a similar manner to raise *cinerea* and *rufibarbis* to specific rank. The constant presence of erect hairs on the gula in the former and the peculiarities of nidification and of temperament in the latter, and the complete absence or extreme rarity of transitions between these forms and *fusca* certainly justify this procedure.

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[1] Der wanderzug der Steppen-u. Wüstenameisen von Zentral-Asien nach Süd-Europa und Nord-Afrika. Zool. jahrb. Suppl., 1912, **15**, p. 95–104. Emery's statement refers only to *F. lasioides*, which in my opinion is merely a subspecies of *neogagates* (*vide infra*).

[2] That the ethological affinities of Proformica and Neoformica with Formica *sens. str.* are extremely close is shown by the fact that such form as *P. neogagates* and *N. incerta* sometimes function, either alone or in company with *F. fusca*, as slaves, or auxiliaries of *F. sanguinea*.

Among the Nearctic and Palaearctic Camponotinae the only genera at all closely related to Formica are Polyergus, Lasius, Myrmecocystus, and Cataglyphis. The parasitic genus Polyergus is now generally believed to have been directly derived from Formica. This may be admitted without accepting Wasmann's more specific assertion that it has arisen from *F. sanguinea,* since there is no morphological basis for this statement but merely the inference that the slave-making habits of Polyergus are in a more advanced or specialized stage of phylogenetic development than those of *sanguinea.* It is, of course, possible that the slave-making habits have been developed independently in the two genera. Lasius has been quite distinct from Formica since Eocene or even Mesozoic times, since we find in the Baltic amber, which is attributed to the Lower Oligocene, a typical Lasius (*L. schiefferdeckeri* Mayr), scarcely distinguishable from small varieties of the existing *L. niger* L., and a species of Formica (*F. flori* Mayr) perhaps identical with the living *F. fusca.* The relations of the genera Myrmecocystus and Cataglyphis have been recently considered by Emery.[1] The Old World species of Cataglyphis were supposed to be congeneric with the American species of Myrmecocystus till a few years ago, when I showed that the New and Old World forms must be at least subgenerically distinct owing to the differences in the males and in the arrangement of the ammochaetae in the workers and females.[2] More recently Emery and Forel have separated them generically, and the former author concludes that the New World Myrmecocystus arose from the genus Lasius, whereas the Old World Cataglyphis was derived from Proformica. If we accept this conclusion Myrmecocystus and Cataglyphis are heterophyletic genera and their similarity is due either to their having arisen from allied genera or to their having converged through adaptation to life in dry, hot deserts.

There has been some discussion between Wasmann and Emery concerning the phylogeny of the species representing various groups of Formica *sens. str.*, notably concerning *fusca, rufa,* and *sanguinea.* Wasmann,[1] starting from purely ethological considerations, has endeavored to show that the *rufa* forms have arisen from *fusca* and have in turn given rise to *sanguinea.* In support of this view he calls attention to *F. flori* of the Baltic amber and its very close resemblance to the living *fusca.* The absence of *rufa* and *sanguinea* in the amber seems to be taken to indicate that they had not yet been evolved from *fusca.* This argument is very specious, but a moment's consideration shows its feebleness for, as Emery has pointed out, the mandibles of the male *flori* are completely edentate like those of the modern *fusca* and of many members of its group, whereas the males of *sanguinea* and of many forms of the *rufa* group have distinctly dentate mandibles. We cannot, therefore, derive these forms from *fusca,* since it would be contrary to phylogenetic methods to assume the re-development of denticles on the vestigial mandibles of the descendant of a form in which the denticles had already completely disappeared in the early Tertiary. There is, in fact, nothing to indicate that *fusca* is the type of the most primitive and ancestral group of Formicae or that it is older than *sanguinea* or *rufa.* Emery may be quite right in supposing that these species are quite as old as *fusca* and that the conditions in the amber may be due to *sanguinea* and *rufa* having their origin in America, Eastern Asia, or the polar regions and not having entered the Baltic region till after the amber fauna had become extinct.[2]

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[1] Der Wanderzug etc. *Loc. cit.*, p. 102.

[2] Honey-ants, with a revision of the American Myrmecocysti. Bull. Amer. mus. nat. hist., 1908, **24**, p. 345.

Our knowledge of the geographical distribution of the North American Formicae has been very imperfect heretofore, owing to the small amount of material which has passed through the hands of myrmecologists. For this reason I have given prominence to the subject in the present paper by citing all or nearly all the localities from which I have seen specimens. These localities are sufficiently numerous, at least in the case of the more common forms, to enable us to form a fairly accurate conception of their geographical range. I could have wished to see more material from British America, from the states of Kentucky, Tennessee, Alabama, and Mississippi and from the mountains of Northern Mexico. These are regions in which, unfortunately, very few ants have been collected.

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[1] Ueber den ursprung des sozialen parasitismus, der sklaverei und der myrmekophilie bei den ameisen. Biol. centralbl., 1909, **29**, p. 587 *et seq.*

[2] Since this paragraph was written I have discovered in addition to *F. flori* four undescribed species of Formica in the Baltic amber. One of these, *F. horrida,* sp. nov. is closely related to *cinerea,* another, *F. phaëthusa,* sp. nov. to *truncicola,* another, *F. clymene,* sp. nov., to *rufa,* and the fourth, *F. strangulata,* sp. nov., in the peculiar structure of its thorax, recalls certain species of Prenolepis (*e. g. P. imparis* Say). I find also that the ant described by Mayr as *Camponotus constrictus* may be more properly regarded as an aberrant Formica. These six species show very clearly that the genus Formica was quite as highly specialized in the early Tertiary of Northern Europe as it is at the present time, and that speculations, like those in which Wasmann has been indulging, are utterly futile and misleading.

Formica, Lasius, Stenamma, and Myrmica are the only circumpolar genera that are confined to the Northern hemisphere. Of these Formica is the most eurythermal, ranging in Europe, Asia, and North America from a latitude of 30° to 60° or 65°, and therefore nearly to the Arctic circle.[1] In altitude the species range from sea-level to above timberline on our loftiest mountains (12,000 to 12,500 ft.). The species of both Lasius and Myrmica are more stenothermal, as they spread neither so far north nor so far south, nor to such altitudes. In passing I may note that I have seen no specimens of Formica from Florida although I have studied many collections of ants from that state. The single species known to me from Mexico (*F. perpilosa*) occurs only on the high plateau. In all probability a few other forms, such as *F. gnava* and *F. pilicornis,* will eventually be found in the same region.

The various species, subspecies, and varieties of Formica differ considerably in habitat. Thus *F. pallidefulva sens. str., moki, pilicornis, perpilosa,* and the various forms of *rufibarbis* are so decidedly xerothermal that they are confined to rather arid portions of the upper and lower austral zones in the southern and southwestern states, whereas the typical *fusca* and its varieties *subaenescens, marcida,* and *gelida, F. sanguinea subnuda* and *F. ulkei* are essentially boreal or subalpine and properly belong to the Canadian and Hudsonian zones. The majority of the species, however, are characteristic insects of Merriam's transition zone.

Practically all of the species find their optimum environment in hilly or mountainous country at moderate elevations, where there are open woods and thickets of deciduous or mixed trees and shrubs, where the rain-fall is abundant and there is nevertheless plenty of heat and sunshine during the summer months, and where, owing to the sloping surface of the soil and abundance of stones, the land is neither flooded nor parched during certain periods of the year. Hence we find the species of Formica most conspicuously abundant and their colonies most numerous and populous in the mountain regions of both continents, notably in the Rockies and Alleghanies, in the Alps, Caucasus, and Ural Mountains. A similar though less pronounced abundance of species and colonies is noticeable in the hilly or rolling portions of the transition zones of both continents, owing to the similar, though somewhat less favorable conditions of temperature, moisture, and vegetation. In more level and arid regions, such as the deserts, the genus Formica is replaced by Myrmecocystus in the New, and Cataglyphis in the Old World.

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[1] Kolbe (Glazialzeitliche reliktenfauna im hohen norden. Deutschr. ent. zeitschr., 1912, p. 33–63) mentions Formica (presumably *F. fusca*) as occurring even as far north as 67° 34′ at Werchojansk on the Jana River in the province of Irkutsk, Siberia. This is said to be one of the coldest spots on the planet, with a minimum temperature of −60° to −67° C.

If we divide the total number of known Formicae (144) into Old and New World forms, we find that Eurasia possesses only fifty-two, whereas North America, though a much smaller land area, possesses ninety-three species, subspecies, and varieties.[1] This would seem to indicate that the latter continent must be the original home of the genus, especially as it possesses representatives of all the Eurasian groups of species besides two peculiar to itself (the *microgyna* group and the subgenus Neoformica). Unless we accept the view that the genus arose in the polar region during Mesozoic times and radiated its species out into Europe, Asia, and North America, we must suppose that Eurasia has received its species by immigration from the Nearctic region. That the latter view is the more probable is shown by a glance at the distribution of the forms in America. At least thirty-nine of our ninety-three forms, or nearly 42%, occur in Colorado and the adjacent portions of New Mexico. Not only are these two states thus abundantly supplied with species, subspecies, and varieties but the colonies of the individual forms are unusually numerous and flourishing on the mountain slopes of this territory. We may therefore regard the southern ranges of the Rocky Mountains in the United States as the center of origin of the genus and of the dispersal of species to other portions of North America.

Formica thus affords striking confirmation of the views of Adams[2] and Scharff[3] that the southwestern states and the adjacent portions of Mexico are the seat of one of the most active North American centers of species formation and dispersal of both plants and animals. It is true that the Formica center does not accurately correspond with the southwestern center as defined by Adams for the biota in general, since the former lies somewhat further north and is much less arid, but this is, perhaps, a matter of minor importance. Both Adams and Scharff recognize another center of species formation and dispersal in the southeastern states, but none of our Formicae seems to have arisen in this region, although this does not apply to other ant-genera. *F. pallidefulva* is the only species of the genus that might be supposed to have originated in such a center, but the occurrence of some of the subspecies of *pallidefulva* as far west as Texas, New Mexico, and Colorado and the existence of an allied species, *F. moki,* in Utah and Arizona are by no means inconsistent with a southwestern origin. There seems to have been some obstacle to the spread of many forms westward from Colorado and New Mexico, for no forms of *rufa* or *sanguinea,* or of the *microgyna* and *pallidefulva* series are known to occur in California.

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[1] One of the species, *F. fusca*, is counted twice, because it occurs in both hemispheres.

[2] The Postglacial dispersal of the North American biota. Biol. bull., 1905. **9**. p. 53–71, 1 fig.

[3] Distribution and origin of life in America. Macmillan Co. 1912.

If we assume that the genus Formica had its origin in a southwestern center, we must conclude that the emigration of species from this region to other parts of North America and especially to Asia over a Bering Sea land-bridge and to Europe across Scharff's Greenland-Iceland land-bridge, has extended over a very long period of time. The first emigrants must have reached the Old World before Oligocene and probably as early as late Mesozoic times, because we find *F. flori* as a common ant in the Baltic amber. Precursors of the *rufa, sanguinea,* and *exsecta* groups must have reached the Old World at the same time or somewhat later. That these various species have since occupied the territory which they invaded, without being dislodged during the glacial epoch is very probable. Both Kolbe[1] and Scharff have recently given good reasons for maintaining that the biogeographical conclusions so generally accepted as following from the statements of those geologists who have asserted the existence of a very extensive and severe glaciation of the northern portions of all the great land masses in the northern hemisphere during the Pleistocene, must be, to a considerable extent, erroneous. These investigators hold that glaciation could not have been so extensive as to have "sterilized" the greater part of North America and Eurasia, but that temperature and other conditions during the Pleistocene must have been sufficiently favorable to admit of the survival of a rather considerable fauna and flora in the immediate neighborhood of the glaciers. Hence many species were able to maintain the station which they had occupied since early Tertiary or Mesozoic times. As it is probable that these views will before long cause a revolution in our biogeographical and geological conceptions, it is timely to call attention to the fact that the boreal distribution of the species of Formica, especially of the typical *F. fusca,* is in complete accord with the views of Kolbe and Scharff. This is also true of certain other ants, *e. g. Camponotus whymperi, Lasius niger* and several of the species of Myrmica.

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[1] Glazialzeitliche reliktenfauna etc. *Loc. cit.*

In order to facilitate the identification of the various species, subspecies, and varieties of Formica, I append dichotomic tables of the worker phases. I have added tables of the females of the *rufa* and *microgyna* groups, because their females are usually much more easily identified than their workers. It is often difficult or impossible to identify isolated Formica workers or specimens that are not perfectly clean and well preserved. For this reason the collection and description of single worker specimens of these ants, as if they were butterflies or beetles, should be discouraged.

KEY TO THE SUBGENERA AND GROUPS.

1. First funicular joint of worker and female about as long as the second and third joints taken together, the latter shorter or at least not longer than the penultimate joints. Frontal carinae short, subparallel, not diverging behind. Stipes of male genitalia much longer than the volsellae and sagittae. Small, mostly smooth, shining, dark-colored species.
Subgenus PROFORMICA Ruzsky.

First funicular joint of worker and female distinctly shorter than the second and third joints taken together, the latter longer than the penultimate joints of the antennae. Stipes of male genitalia but slightly longer than the volsellae and sagittae except in the subgenus Neoformica.
2. Subgenus FORMICA Linné.

2. Anterior border of clypeus of worker and female, and often also of the male, notched or emarginate in the middle.
sanguinea group.

Anterior border of clypeus of worker, female and male entire, rounded or subangularly produced in the middle..........3.

3. Sides of head subparallel, posterior border deeply and broadly excised in the worker and female and often also in the male. Basal border of mandibles with vestiges of denticles.
exsecta group.

Sides of head of worker usually converging anteriorly, posterior border of head of worker and female straight or convex or at most very feebly excised. Basal border of mandibles without vestiges of teeth....................................4

4. Body of worker robust. Head of largest individuals not or scarcely longer than broad. Funicular joints 2–3 longer and more slender than joints 6–8. Petiole usually with rather sharp border. Body opaque, color of species light or dark red with brown or black gaster....................................5

Body of worker more slender. Head of largest individuals usually distinctly longer than broad. Funicular joints 2–3 only slightly more slender than joints 6–8. Petiole usually narrow, rather thick and with blunt border. Color and sculpture diverse....................................6

5. Female larger than the largest workers, measuring 6–11 mm. *rufa* group.

Females not larger and sometimes even smaller than the large workers, measuring only 4–6 mm...........*microgyna* group.

6. Thorax of worker rather short. Median joints of funiculi usually less than 1½ times as long as broad; scapes stout, distinctly curved at the base. Petiole flattened behind. Stipes of male genitalia but slightly longer than the volsellae and sagittae. *fusca* group.

Thorax of worker longer. Median joints of funiculi more than 1½ times as long as broad; scapes slender, scarcely curved at the base. Petiole convex behind. Stipes of male genitalia much longer than the volsellae and sagittae. Subgenus NEOFORMICA, subgen. nov.

SUBGENUS FORMICA.

SANGUINEA GROUP.

Workers.

1. Palaearctic forms....................................2

Nearctic forms....................................6

2. Head and thorax rich red or brownish red, head above more or less infuscated....................................3

Head and thorax more yellowish red, head above not infuscated.5

3. Infuscation of head extending down onto the cheeks, leaving only the posterior corners red . . . . *sanguinea fusciceps* Emery.
Infuscation of head less extensive, confined to the front and vertex. 4
4. Epinotum obtusely but distinctly angular. . *sanguinea* Latreille.
Epinotum much rounded. . . . . *sanguinea* var. *mollesonae* Ruzsky.
5. Eyes of the usual size and shape. . *sanguinea* var. *clarior* Ruzsky.
Eyes smaller and more elongate. . *sanguinea* var. *flavorubra* Forel.
6. Gaster red or ferruginous like the head and thorax.
bradleyi, sp. nov.
Gaster brown or black, always darker than the head and thorax . 7
7. Gaster decidedly shining, with very sparse, short pubescence. . 8
Gaster opaque or subopaque, with longer, dense pubescence. . . 9
8. Erect hairs on head, thorax, and gaster long and dense. Clypeal notch indistinct in small workers. Length of worker 3.5–5 mm., of female 7.5–9 mm. . . . . *perpilosa* Wheeler.
Erect hairs on head, thorax, and gaster shorter and sparser. Clypeal notch distinct in small workers. Length of worker 3.5–4.5 mm.; of female 6–7 mm. . . . . *manni,* sp. nov.
9. Head, thorax, and petiole brownish testaceous; cheeks straight or slightly concave. 10
Head, thorax, and petiole red or ferruginous; cheeks more or less convex. 11
10. Head long and narrow; antennae slender, scapes not thickened towards their tips; body subopaque. . . . . *pergandei* Emery.
Head shorter; antennae more robust; scapes slightly thickened towards their tips; erect hairs less numerous. . *emeryi,* sp. nov.
11. Hairs on the dorsal parts of the body abundant, conspicuous, glistening white, obtuse or clavate. 12
Hairs less abundant and more slender. 13
12. Head and thorax deep red, petiole infuscated; body slender, mesoëpinotal constriction shallow, epinotum long and low.
munda Wheeler.
Head, thorax, and petiole yellowish red, body stout, mesoëpinotal constriction deep, epinotum short and high.
sanguinea obtusopilosa Emery.
13. Front and vertex more or less infuscated. . *sanguinea aserva* Forel.
Front and vertex not infuscated. 14
14. Hairs nearly always absent on the thoracic dorsum and petiolar border, short and few on the head and gaster.
sanguinea subnuda Emery.

Hairs present on thoracic dorsum, longer and more numerous on head and gaster 15

15. Gaster black 16

Gaster brown 17

16. Body rather opaque; petiole broad, with sharp superior border. *sanguinea rubicunda* Emery.

Body somewhat shining; petiole narrower, with blunter superior border. . . . . *sanguinea rubicunda* var. *lucidula*, var. nov.

17. Sides of head convex; clypeal notch shallow; hairs moderately abundant; tibiae with very fine appressed pubescence 18

Sides of head very feebly convex; clypeal notch rather deep; hairs more abundant; anterior surfaces of tibiae with small oblique hairs and without appressed pubescence. *sanguinea puberula* Emery.

18. Head, thorax, and petiole red, gaster dark brown. Clypeal notch feeble but distinct. . . . . *sanguinea subintegra* Emery.

Head, thorax, and petiole yellow, gaster pale brown. Clypeal notch obsolescent. . *sanguinea subintegra* var. *gilvescens*, var. nov.

RUFA GROUP.

Workers.

1. Palaearctic forms 2

Nearctic forms 12

2. Frontal area opaque; antennal scapes short and robust; whole upper surface of head black. . . . . *uralensis* Ruzsky.

Frontal area shining; antennal scapes longer and more slender; infuscation of head, when present, less extensive 3

3. Gaster brown or black, more or less reddish at the base 4

Gaster entirely black 7

4. Front, vertex, and a small pronotal spot dark brown or blackish; erect hairs on head and thorax usually sparse; eyes hairless . .5

Front, vertex, and pronotum rarely spotted; body and legs usually more hairy; eyes usually hairy 8

5. Head long and narrow, with straight cheeks; thorax slender *rufa* var. *santschii*, nom. nov.

Head short, cheeks more convex; thorax robust 6

6. Head, thorax, and petiole red. *rufa* Linné and its var. *rufopratensis* Forel.

Head, thorax, petiole, and legs brown; hairs sparser.
rufa var. *meridionalis* Ruzsky.

7. Head, thorax, and petiole red; black spots on head and thorax large, those on the pro- and mesonotum confluent.
rufa pratensis Retzius.

Head, thorax, petiole, and legs darker and more brownish; black on thorax more extensive. . . *rufa pratensis* var. *nigricans* Emery.

8. Flexor surfaces of tibiae with numerous erect or suberect hairs . . 9

Flexor surfaces of tibiae without erect hairs 11

9. Color of head, thorax, and petiole bright, yellowish red 10

Color and pilosity transitional to *pratensis*.
rufa pratensis var. *truncicolo-pratensis* Forel.

10. Hairs on head and thorax abundant; eyes hairy; gaster dark brown, with red basal spot . . . . *truncicola* Nylander.

Hairs absent on head and thorax; gaster opaque, black, with red basal spot . . . . *truncicola dusmeti* Emery

11. Bright red; upper surface of head and clypeus hairy.
truncicola var. *yessensis* Forel.

Deep, dull red; upper surface of head and clypeus without hairs.
truncicola var. *sinensis*, var. nov.

12. Antennal scapes with erect hairs 13

Antennal scapes without erect hairs 14

13. Head and thorax bright yellowish red; legs reddish brown.
oreas Wheeler.

Red portions of body darker; legs dark brown; erect hairs on all parts of the body more abundant, shorter on gaster.
oreas var. *comptula*, var. nov.

14. Cheeks and posterior corners of head very convex and rounded; petiole narrow below when seen from behind, broadened above, with straight transverse superior border 15

Cheeks and posterior corners of head less convex and rounded, petiole not transversely truncated above 17

15. Erect hairs absent on gula and upper surface of head, thorax, and petiole 16

Erect hairs present on gula and upper surface of head, thorax, and petiole . . . . *dakotensis* Emery var. *montigena* Wheeler.

16. Gaster black or very dark brown; pubescence on head and thorax very short and indistinct . . . . *dakotensis* Emery.

Gaster paler; pubescence longer and more distinct on head and thorax . . . . *dakotensis* var. *specularis* Emery.

17. Frontal area opaque . . . . *foreliana*, sp. nov.

Frontal area smooth and shining 18

18. Petiole narrow and very low, its border very blunt and not produced upward in the middle.........*ferocula*, sp. nov.
Petiole broader and higher, its border sharp, more or less produced upward in the middle..........................19

19. Erect hairs absent on gula and upper surface of head and thorax. 20
Erect hairs present on gula and upper surface of head and thorax. 22

20. Small forms (4–6.5 mm.)...............*criniventris* Wheeler.
Larger forms (4–9 mm.).................................21

21. Gaster black, somewhat shining, with short, sparse pubescence. *truncicola integra* Nylander.
Gaster dark brown, opaque, densely gray pubescent. *truncicola integroides* Emery var. *haemorrhoidalis* Emery.

22. Eyes hairless..23
Eyes hairy...26

23. Erect hairs on gaster very numerous, very short and stubby... 24
Erect hairs on gaster less numerous, longer.................25

24. Gaster blackish brown .... *comata* Wheeler.
Gaster reddish brown.........................*ciliata* Mayr.

25. Gaster dark brown, opaque, densely gray pubescent. *truncicola mucescens*, subsp. nov.
Gaster black, somewhat shining, finely and sparsely pubescent. *truncicola obscuriventris* var. *gymnomma* Wheeler.

26. Head and thorax of small workers decidedly darker than in largest workers.....................................27
Head and thorax of small workers scarcely or not at all darker than in largest workers...............................30

27. Flexor surfaces of tibiae with erect hairs..................28
Flexor surfaces of tibiae without erect hairs...............29

28. Thorax of large workers bright red like the head or at most very feebly infuscated; pubescence on gaster dense. *rufa aggerans* Wheeler.
Thorax of large workers deeply infuscated; pubescence on gaster more dilute......*rufa aggerans* var. *melanotica* Emery.

29. Head and thorax of large workers entirely or almost entirely without dark spots................*rufa obscuripes* Forel.
Front, vertex, occiput, and thoracic dorsum of large workers blackish .... *rufa obscuripes* var. *whymperi* Forel.

30. Gaster brown, opaque, densely gray pubescent..............31
Gaster black, feebly shining, sparsely and finely pubescent. *truncicola obscuriventris* Mayr.

31. Erect hairs sparse on upper surface of head, thorax, and petiole. *truncicola integroides* Emery.
Erect hairs on upper surface of head, thorax, and petiole dense and abundant. *truncicola integroides* var. *coloradensis*, var. nov.

Females.

1. Palaearctic forms....................................2
Nearctic forms....................................5
2. Frontal area opaque; antennal scapes short and stout. *uralensis* Ruzsky.
Frontal area shining; antennal scapes longer and more slender..3
3. Gaster very smooth and shining, scarcely pubescent..*rufa* Linné.
Gaster opaque or subopaque, distinctly pubescent...........4
4. Gaster opaque, densely pubescent, brownish black, except the extreme base and tip, which are reddish...*rufa pratensis* Retzius.
Gaster subopaque, brown with red base, or red with fuscous posterior margins to the segments.......*truncicola* Nylander.
5. Antennal scapes with numerous erect or suberect hairs......6
Antennal scapes without erect hairs, or with only a few on the posterior surfaces....................................7
6. Hairs on scapes and legs oblique or suberect....*oreas* Wheeler.
Hairs on scapes and legs more erect; on the body coarser and more abundant...............*oreas* var. *comptula*, var. nov.
7. Gaster invested with very long, appressed hairs............8
Gaster not invested with such hairs......................10
8. Gaster yellowish red like the head and thorax, its long, appressed hairs hooked or curved at their tips......................9
Gaster blackish brown, except the base and anal region; its long, appressed hairs not curved at their tips...*comata* Wheeler.
9. Petiolar border with a fringe of very long hairs....*ciliata* Mayr.
Petiolar border without a fringe of long hairs. *criniventris* Wheeler.
10. Head and thorax very smooth and shining, petiole with transversely truncated superior border.....................11
Head and thorax opaque or subopaque, petiole not truncated above.................................... 13
11. Gaster brown....................................12
Gaster red with brown posterior borders to the segments. *dakotensis* var. *specularis* Emery.

12. Mesonotum immaculate.................. *dakotensis* Emery.
Mesonotum with three dark spots.
dakotensis var. *montigena* Wheeler.
13. Gaster very smooth and shining, with very short, sparse, indistinct pubescence....................................14
Gaster opaque or subopaque, with denser, longer pubescence..16
14. Length 7–8 mm. Head and thorax deep red, the former not infuscated. Wings deeply infuscated at the base.
truncicola obscuriventris Mayr.
Length 8–9 mm. Head and thorax dull, yellowish red, both more or less infuscated. Wings only slightly infuscated....15
15. Posterior border of pronotum infuscated.
rufa aggerans, nom. nov.
Pronotum and other portions of thorax more extensively infuscated................ *rufa aggerans* var. *melanotica* Emery.
16. Head and thorax without erect hairs above.................17
Head and thorax with erect hairs above...................19
17. Length 6.5–8 mm. Ground color of head, thorax, and petiole sordid brownish yellow, gaster blackish brown, not paler at the base.................. *truncicola mucescens*, subsp. nov.
Length 8–10 mm. Ground color of head, thorax, and petiole bright or deep red, base of gaster red....................18
18. Pubescence on gaster rather sparse and short, so that its surface is subopaque. . *truncicola integroides* var. *haemorrhoidalis* Emery.
Pubescence on gaster longer and denser so that its surface is more opaque............................ *truncicola integra* Nyl.
19. Hairs on upper surface of head and thorax sparse; wings opaque gray......................... *truncicola integroides* Emery.
Hairs on upper surface of head and thorax somewhat more abundant; wings distinctly infuscated at the base.
truncicola integroides var. *coloradensis*, var. nov.

MICROGYNA GROUP.

Workers.

1. Gaster shining, very sparsely and finely pubescent............2
Gaster opaque, densely pubescent.........................3
2. Head and thorax deep red, gaster black..... *nepticula* Wheeler.
Head and thorax reddish yellow, gaster brown. . *morsei* Wheeler.

3. Antennal scapes with erect or suberect hairs................4
Antennal scapes without erect or suberect hairs..............5
4. Hairs on antennal scapes coarse and clavate...*impexa* Wheeler.
Hairs on antennal scapes delicate, not clavate.
microgyna Wheeler.
5. Border of petiole blunt; head and thorax rich yellowish red...6
Border of petiole sharp and compressed; head and thorax sordid brownish red..10
6. Gaster black or dark brown, not red at the base..............7
Gaster brown, red at the base.................................9
7. Largest workers with the pro- and mesonotum and also the ocellar region infuscated.
microgyna rasilis var. *spicata*, var. nov.
Largest workers without the head and thorax infuscated or at most with dark ocellar triangle..........................8
8. Tibiae with abundant, short, subappressed hairs on their extensor surfaces...........*microgyna* var. *recidiva*, var. nov.
Tibiae without such hairs.........*microgyna rasilis* Wheeler.
9. Erect hairs on head, thorax, and gaster moderately numerous, usually lacking on posterolateral corners of head.
difficilis Emery.
Erect hairs more abundant and longer, especially on the front, gula, and thorax, present on posterolateral corners of head.
difficilis var. *consocians* Wheeler.
10. Posterior portion of head, a spot on the pronotum and one on the mesonotum dark brown or blackish......*adamsi* Wheeler.
Infuscation of head and thorax more restricted, frontal area smoother and more shining......*adamsi* var. *alpina* Wheeler.

Females.

1. Gaster reddish yellow like the head and thorax..............2
Gaster brown or black.......................................3
2. Tibiae without long, oblique hairs............*difficilis* Emery.
Tibiae with long, oblique hairs..*difficilis* var. *consocians* Wheeler.
3. Gaster smooth and more or less shining, finely and sparsely pubescent.......................................4
Gaster opaque, with denser, longer pubescence..............6
4. Antennal scapes without erect hairs..*microgyna scitula*, subsp. nov.
Antennal scapes with erect hairs...........................5

5. Antennal scapes with very few erect hairs. . *nepticula* Wheeler.
Antennal scapes with numerous erect hairs. . . *nevadensis* Wheeler.
6. Antennal scapes with erect hairs. 7
Antennal scapes without erect hairs. 8
7. Hairs on antennal scapes coarse and clavate. . . . *impexa* Wheeler.
Hairs on antennal scapes delicate, not clavate.
microgyna Wheeler.
8. Clavate hairs on head, thorax, and gaster rather short.
microgyna rasilis Wheeler.
Clavate hairs on head, thorax, and gaster longer.
microgyna rasilis var. *spicata*, var. nov.

EXSECTA GROUP.

Workers.

1. Nearctic forms. 2
Palaearctic forms. 6
2. Antennal scapes thickened towards their tips. 3
Antennal scapes not thickened towards their tips. 5
3. Posterior half of head black. . . . . *ulkei* Emery.
Posterior half of head brown or red. 4
4. Gaster brown, subopaque or slightly shining.
ulkei var. *hebescens*, var. nov.
Gaster black, more opaque and pubescent.
exsectoides opaciventris Emery.
5. Petiole thick and narrow, with sharp but not cultrate border, transversely truncated. . . . . . *exsectoides* var. *hesperia*, var. nov.
Petiole thin, broad, with a sharp, cultrate border which is not transversely truncated
exsectoides Forel and var. *davisi* Wheeler.
6. Head long, its posterior border deeply excised; body opaque or but feebly shining. 7
Head shorter, its posterior border less deeply excised; body more shining. . . . . *suecica* Adlerz.
7. Clypeus without a transverse impression behind its anterior border; maxillary palpi long. 8
Clypeus with a transverse impression behind its anterior border; maxillary palpi short. 10

8. Petiole broad, its superior border rounded, entire or only slightly excised in the middle............*exsecta* var. *etrusca* Emery.

Petiole narrow, its superior border deeply excised in the middle.9

9. Red or sordid yellowish red, posterior portion of head and a large spot on the pronotum brown..........*exsecta* Nylander.

Body paler red, vertex and pronotum each with a small brown spot...........................*exsecta* var. *rubens* Forel.

10. Gaster with a red spot at the base. *exsecta pressilabris* var. *rufomaculata* Ruzsky.

Gaster without a red spot at the base......................11

11. Gaster, especially at the base, lustrous *exsecta pressilabris* Nylander.

Base of gaster opaque, pubescence somewhat longer. *exsecta pressilabris* vars. *foreli* Emery and *exsectopressilabris* Forel.

FUSCA GROUP.

Workers.

1. Palaearctic forms.......................................2

Nearctic forms...15

2. Gula without erect hairs................................3

Gula with erect hairs..................................13

3. Body opaque or subopaque........................... 4

Body shining..12

4. Thorax black, like the head and gaster, in both large and small workers.. 5

Thorax, at least in the large workers, largely red..............7

5. Body dull and opaque, rather coarsely shagreened. *fusca* var. *japonica* Motschulsky.

Body subopaque, more finely shagreened..................6

6. Pubescence of gaster short, delicate, not silky.....*fusca* Linné.

Pubescence of gaster, long, silky; head, legs, scapes, and sutures of thorax reddish or yellowish...*fusca* var. *glebaria* Nylander.

7. Largest workers only with red thorax..*fusca* var. *rubescens* Forel.

Thorax rarely infuscated except in small workers............8

8. Erect hairs lacking or very sparse; pubescence delicate and not very dense; red portions of body pale.....................9

Erect hairs more abundant; pubescence denser, red portions of body darker..10

9. Body opaque, distinctly shagreened. . *rufibarbis* var. *clara* Forel.
Body subopaque, lustrous, finely shagreened.
rufibarbis var. *caucasica* Ruzsky.

10. Erect hairs yellow: pubescence of gaster without silky luster.
rufibarbis Fabricius.
Erect hairs whitish, pubescence on gaster longer. 11

11. Gaster with a bluish tinge. . . . .*rufibarbis* var. *glauca* Ruzsky.
Gaster gray, red of body somewhat paler.
rufibarbis var. *subpilosa* Ruzsky.

12. Length 3–6.5 mm. Epinotum angular in profile, body slender.
fusca picea Nylander and its var. *gagatoides* Ruzsky.
Length 5–7.5 mm. Epinotum rounded in profile, body stout.
gagates Latreille and its var. *fuscogagates* Forel.

13. Body slender, thorax long, with very long, shallow, saddle-shaped mesoëpinotal constriction. . . . .*subrufa* Roger.
Body stouter, thorax of the usual shape. 14

14. Body dark brown or blackish.
cinerea Mayr and its vars. *fuscocinerea* Forel and *armeniaca* Ruzsky.
Body light reddish brown. . . . . *cinerea* var. *imitans* Ruzsky.

15. Gula without erect hairs. 16
Gula with erect hairs. 26

16. Gaster opaque or subopaque, densely pubescent. 17
Gaster more shining, very sparsely pubescent. 24

17. Thorax black or very dark brown. 18
Thorax largely red. 21

18. Pubescence on gaster short, not silky. . . . .*fusca* Linné.
Pubescence on gaster longer, denser and silky. 19

19. Body black, pubescence not silvery. .*fusca* var. *subsericea* Say.
Body dark brown. 20

20. Pubescence on body not silvery, sutures of thorax reddish or yellowish. . . . .*fusca* var. *marcida*, var. nov.
Pubescence on body somewhat longer, denser, and silvery.
fusca var. *argentea* Wheeler.

21. Gaster black or blackish brown; epinotum angular in profile. . .23
Gaster reddish brown, paler, epinotum rounded in profile. . . .22

22. Gaster more or less infuscated above; length 3–6 mm.
fusca var. *neoclara* Emery.
Gaster not infuscated above, length 3–3.5 mm.
fusca var. *blanda*, var. nov.

23. Length 4–7.5 mm.; gaster opaque, with long, dense pubescence.
rufibarbis var. *occidua* Wheeler.

Length 3.5–6 mm.; gaster somewhat bronzy, slightly shining, with shorter pubescence......*rufibarbis* var. *gnava* Buckley.

24. Thorax entirely black..........*fusca* var. *subaenescens* Emery.
Thorax more or less red..................................25

25. Thorax clear, yellowish red throughout. *fusca* var. *neorufibarbis* Emery.
Thorax of large workers infuscated or black anteriorly. *fusca* var. *gelida*, var. nov.

26. Body opaque or subopaque, head of largest workers not rectangular.. .28
Body shining, head of largest workers rectangular............27

27. Thorax brownish red or dark chestnut......*subpolita* Mayr.
Thorax yellow or yellowish brown, head of largest workers with more nearly parallel sides. *subpolita* var. *camponoticeps*, var. nov.

28. Antennal scapes with erect hairs, eyes densely hairy. *cinerea pilicornis* Emery.
Antennal scapes without erect hairs, eyes not hairy..........29

29. Erect hairs abundant on head and thorax; length 3.5–6 mm...30
Erect hairs very sparse on head and thorax; length 5–6.5 mm. *sibylla*, sp. nov.

30. Gaster blackish or dark brown; frontal area opaque..........31
Gaster pale reddish brown, not infuscated above; frontal area shining.................................*montana* Emery.

31. Petiole broad, seen from behind cordate, notched in the middle. *cinerea* var. *altipetens*, var. nov.
Petiole narrower, with blunt margin, usually entire or obtusely angular in the middle................................32

32. Suberect hairs absent on sides of head and flexor surfaces of legs. 33
Suberect hairs present on sides of head and flexor surfaces of legs. *cinerea* var. *lepida*, var. nov.

33. Body dark brownish, top of head and gaster blackish. *cinerea* var. *neocinerea* Wheeler.
Body light yellowish red, top of head, pronotum, and gaster brown.....................*cinerea* var. *rutilans*, var. nov.

SUBGENUS PROFORMICA.

Workers.

1. Palaearctic forms..2
Nearctic forms..8

2. Gula with long curved hairs (ammochaetae); maxillary palpi long . . . . *emmae* Forel.
Gula without ammochaetae, palpi shorter 3
3. Antennal scapes with suberect hairs . . . . *aberrans* Mayr.
Antennal scapes without suberect hairs 4
4. Erect hairs on body short and clavate . . . . *kraussi* Forel.
Erect hairs on body longer, not clavate 5
5. Head long and narrow, especially in small workers, thorax slender. 6
Head broader, antennae shorter and thicker, thorax stouter. *mongolica* Emery.
6. Whole body opaque, densely pubescent . . . . *korbi* Emery.
At least the gaster very smooth and shining, pubescence very dilute or absent 7
7. Only the front and clypeus finely longitudinally striated. *nasuta* Nylander.
Head striated nearly as far back as the occiput. *nasuta* var. *striaticeps* Forel.
8. Antennal scapes with erect hairs 9
Antennal scapes without erect hairs 10
9. Body yellowish brown, gaster and posterior portion of head darker . . . . *neogagates lasioides* Emery.
Body black or very dark brown, thorax sometimes piceous or reddish . . . . *neogagates lasioides* var. *vidua* Wheeler.
10. Erect hairs on dorsal surface of body abundant; body moderately shining 11
Erect hairs on body very sparse or absent; body very smooth and shining . . . . *limata,* sp. nov.
11. Erect hairs on body very delicate . . . . *neogagates* Emery.
Erect hairs on body coarser. *neogagates* vars. *morbida,* var. nov. and *vinculans,* var. nov.

Subgenus Neoformica.

Workers.

1. Body opaque . . . . *moki* Wheeler.
Body shining 2
2. Erect hairs present on gula and petiole 3
Erect hairs absent on gula and petiole 5

3. Gaster distinctly infuscated, darker than the head and thorax. .4
Gaster scarcely darker than the head and thorax, its pubescence longer and denser. . *pallidefulva schaufussi* var. *dolosa* Wheeler.
4. Hairs on gula and petiole numerous and conspicuous.
pallidefulva schaufussi Mayr.
Hairs on gula and petiole few, often lacking on one or the other; head, thorax, and gaster darker.
pallidefulva schaufussi var. *incerta* Emery.
5. Gaster yellow like the head and thorax or but very slightly infuscated. 6
Gaster dark brown or blackish, head and thorax light or dark brown or reddish. 7
6. Body pale yellow. . . . . *pallidefulva* Latreille.
Body reddish yellow, pubescence on gaster shorter, whole body smoother. . . . . *pallidefulva* var. *succinea* Wheeler.
7. Head and thorax brown or reddish, gaster shining.
pallidefulva nitidiventris Emery.
Head and thorax darker, body often less shining.
pallidefulva nitidiventris var. *fuscata* Emery.

SUBGENUS FORMICA (Linné) Ruzsky.

Sanguinea Group.

1. FORMICA SANGUINEA SANGUINEA Latreille.

Formica sanguinea Latreille, Essai hist. fourmis France, 1798, p. 37, ☿; Hist. nat. fourmis, 1802, p. 150, pl. 5, fig. 29, ☿; Lepeletier, Hist. nat. insect. Hymén., 1836, **1**, p. 203, ☿ ♀ ♂; Förster, Hymen. stud., 1850, **1**, p. 20; Mayr, Verh. Zool. bot. ver. Wien, 1855, **5**, p. 336; Nylander, Ann. sci. nat. Zool., 1856, ser. 4, **5**, p. 62; F. Smith, List Brit. anim. Brit. mus., 1858, pt. 6, p. 115; Mayr, Europ. Formicid., 1861, p. 46–48; Forel, Denks. Schweiz. gesell. naturw., 1874, **26**, *passim;* Lubbock, Journ. Linn. soc. Zool., 1877, **13**, p. 217, pl. 17, fig. 2; Ern. André, Spec. Hymén. Europe, 1882, **2**, pt. 14, p. 180, p. 185, 188, pl. 9, fig. 18; Dalla Torre, Catalog. Hymen., 1893, **7**, p. 211; Bingham, Fauna Brit. Ind., 1903, **2**, p. 336; Ruzsky, Formicar. Imper. Ross., 1905, p. 411, figs. 76–79; Emery, Deutsch. ent. zeitschr., 1909, p. 182.
Formica dominula Nylander, Acta Soc. Fennica, 1846, **2**, p. 905, ☿ ♀ ♂, pl. 18, fig. 15, p. 1047; Ibid., 1849, **3**, p. 26.

WORKER. Length 6–9 mm.

Body robust; head, excluding the mandibles, about as broad as long, narrowed in front, with rather straight sides and feebly and broadly excised posterior border. Mandibles broad. Clypeus with a distinct notch in the middle of its anterior border. Pro- and mesonotum in profile slightly depressed; mesoëpinotal impression rather deep and angular; base and declivity of epinotum forming together a rounded angle. Petiole broad, with a sharp border, which is either entire or with a median emargination.

Body subopaque; head slightly lustrous; mandibles finely striatopunctate. Frontal area not shining.

Hairs whitish, sparse, suberect, present on the dorsal surface of the head, pronotum, fore coxae, and gaster, absent on the border of the petiole; short on the upper surface of the gaster, longer and more abundant on the venter. Legs without hairs, except the row of oblique bristles along the flexor surface of the tibiae and metatarsi. Pubescence very fine and sparse on the head and thorax, longer, dense, and grayish on the gaster.

Head, thorax, petiole, legs, and antennae light or dark red; front and vertex more or less infuscated. Gaster black, with the anus and usually a spot at the base of the first segment, red.

FEMALE. Length 9–11 mm.

Color darker and more brownish than that of the worker; head above and behind black; mandibles, clypeus, cheeks, and antennae brown; mesonotum with an anteromedian, and a pair of parapsidal blotches of the same color. Wings brownish, darker towards the base. Pilosity and pubescence as in the worker.

MALE. Length 7–10 mm.

Mandibles broad, with 4–5 teeth. Clypeus convex, carinate, with a sinuous emargination in the middle of its anterior border. Head, excluding the mandibles, a little broader than long, with straight posterior border and rounded posterior angles, much narrower in the region of the cheeks, which are shorter than half the eyes and feebly concave. Petiole broad and thick below, with thin, sharp superior border, broadly and rather deeply excised in the middle.

Surface of head and thorax opaque, densely shagreened; gaster somewhat glossy.

Hairs short and sparse; pubescence fine, rather uniform on the head, thorax, and gaster, but not dense enough to conceal the surface.

Black; legs yellow; antennae brown or blackish; scapes sometimes paler; genitalia brownish or reddish yellow. Wings colored like those of the female.

HOSTS (SLAVES): *F. fusca; F. fusca* var. *glebaria, F. fusca gagates, F. rufibarbis,* and *F. cinerea.*

According to Emery this, the typical form of the species, is distributed throughout the Palaearctic region, but in the southern portions of Europe and Asia occurs only in hilly or mountainous country. In Europe it ranges south as far as Sicily, in Asia as far as the Himalayas (Cashmir) and Lahoul, on the frontier of Thibet.

The workers of *sanguinea* colonies make raids during the summer months on colonies of *F. fusca* and the other species cited above and pillage their pupae. Many of these are devoured, but a number of them are permitted to develop to maturity in the *sanguinea* nests and thus become "slaves," or "auxiliaries." The colonies are therefore said to be of the "mixed" type. When old, however, these colonies often lose the predatory habit and become slaveless. Viehmeyer, Donisthorpe, and Wasmann have shown that the female *sanguinea* establishes her colony by entering a *fusca* nest, appropriating some of the pupae and killing or driving away any of the *fusca* workers that venture to attack her or seek to deprive her of her booty. She guards the kidnapped young and eventually helps them to hatch, thereby surrounding herself with a troop of nurses for her own brood as soon as she begins to lay. This method of colony formation in the typical *sanguinea* is the same as that first described by myself for our American subspecies *rubicunda* and *subintegra* (*vide* p. 408).

The nests of *sanguinea* have the form of low, obscure mounds of earth, or are excavated under stones or logs or around stumps or the roots of plants, and their openings are often banked with a small amount of vegetable detritus. This ant is restless and fond of moving to new quarters from time to time. In some countries it regularly occupies nests in sheltered situations such as woodlands during the winter months but moves to nests in sunny, open places during the summer. When moving to new nests the workers carry the slaves in their mandibles. The worker *sanguinea* is very courageous and fiercely resents interference with its nests, using its mandibles and injecting formic acid into the wounds made with these.

2. F. SANGUINEA SANGUINEA var. MOLLESONAE Ruzsky.

F. sanguinea var. *mollesonae* Ruzsky, Rev. Russe entom., 1903, p. 206, ☿; Formicar. Imper. Ross. 1905, p. 420; Emery, Deutsch. ent. zeitschr., 1909, p. 184.

WORKER. Differs from the worker of the typical form in having the epinotum much more rounded in profile.

Transbaikalia, Siberia.

3. F. SANGUINEA SANGUINEA var. CLARIOR Ruzsky.

F. sanguinea var. *clarior* Ruzsky, Formicar. Imper. Ross., 1905, p. 420, ☿; Emery, Deutsch. ent. zeitschr., 1909, p. 184.

WORKER. Differs from the typical form in having the red portions of the body paler and the red basal and apical spots of the gaster more pronounced.

Caucasus.

4. F. SANGUINEA SANGUINEA var. FLAVORUBRA Forel.

F. sanguinea var. *flavorubra* Forel, Ann. Soc. ent. Belg., 1909, **53**, p. 105, ☿.

WORKER. "Differs from the typical *sanguinea* in its light and vivid red color, which is somewhat yellowish, clearer, and more yellow than in *F. truncicola* Nyl. The base of the first gastric segment is also more or less yellowish red, and the eyes are a little smaller and slightly more elongate.

Ronda, Andalusia" (*Forel*).
HOST (SLAVE). Probably *F. fusca* var. *glebaria.*

5. F. SANGUINEA SANGUINEA var. FUSCICEPS Emery.

F. sanguinea var. *fusciceps* Emery, Zool. jahrb. Syst., 1895, **8**, p. 335, *nota*, ☿: Deutsch. ent. zeitschr., 1909, p. 184.

WORKER. Red color darker than in the typical form; the blackish brown spot on the vertex extending laterally as far as the eyes and leaving only a small red area on each of the posterior corners of the head.

HOST (SLAVE). Probably *F. fusca* var. *japonica.*
Japan: Yokohama.

6. F. SANGUINEA ASERVA Forel.

.*F sanguinea* st. *aserva* Forel, Ann. Soc. ent. Belg., 1901, **45**, p. 395, ☿ ♀; Wheeler, Bull. Amer. mus. nat. hist., 1906, **22**, p. 85; 1908, **24**, p. 631; Ants, 1910, p. 458, 570.

WORKER. Length 4–7 mm.

Closely related to the typical European form. Clypeal notch rather shallow; clypeal carina more distinct and the surface of the clypeus more convex. Mesoëpinotal constriction a little shallower than in the typical *sanguinea.* Antennal scapes slender at the base, somewhat enlarged towards their tips. Head relatively large in large workers, almost broader than long, excluding the mandibles, with convex sides, rounded posterior corners and straight or feebly excised posterior border. Petiole broad, with sharp, entire or feebly excised superior border.

Sculpture a little finer than in the typical *sanguinea;* head and gaster more shining; punctures on the occiput rather distinct, scattered.

Hairs yellowish, very sparse, usually absent on the thorax and petiole; short on the gaster. Pubescence shorter and more dilute than in the typical *sanguinea,* so that the surface, especially that of the gaster, appears more shining; very fine and appressed on the legs and scapes.

Color brownish red like that of deeply colored specimens of the typical form; posterodorsal portion of head and often also the middle of the pronotum infuscated or blackened.

FEMALE. Length 7–8 mm.

Sculpture, pilosity, and color as in the worker; metanotum, posterior border of pronotum, and scutellum and three spots on the mesonotum dark brown or black. Head, excluding the mandibles, as broad as long, broader behind than in front, with straight posterior and lateral borders. Scale of petiole broad, much compressed anteroposteriorly, with thin, sharp, entire or feebly emarginate border. Wings colored as in the typical *sanguinea.*

MALE. Length 8–8.5 mm.

Mandibles broad, dentate. Clypeus convex, carinate; emargination of its anterior border feeble but distinct. Petiole thick, with rather sharp, transverse border. Hairs and pubescence very short and sparse, so that the thorax and gaster are more shining than in the typical *sanguinea;* head including the mandibles, opaque. Body and antennae black; tips of mandibles brownish; legs brownish yellow. Genitalia rather deeply infuscated. Wings colored like those of the female.

HOSTS (TEMPORARY). *F. fusca* and its var. *subsericea.*

TYPE LOCALITY.— Ontario: Toronto, (Forel).

Nova Scotia: Round Hill; Clark's Harbor, Cape Sable Island (A. Halkett).

New Brunswick: Grand Manan (Centr. Exper. Farms Coll.).

Maine: South Harpswell (Wheeler).

New Hampshire: Franconia (Mrs. A. T. Slosson); Summit of Mt. Washington (C. S. Bacon and Mrs. A. T. Slosson).

Massachusetts: Mt. Wachusett (A. C. Burrill).

Connecticut: Colebrook, 1,500 ft. (Wheeler).

Michigan: Isle Royale (O. McCreary).

Wisconsin: White Fish Bay, near Milwaukee (Wheeler); Beaver Lake (C. E. Brown).

Illinois: Rockford (Wheeler).

This subspecies, in color and pilosity at least, is more closely related to the typical European *sanguinea* than is any of the other North American forms. The distribution shows that it is an essentially boreal ant. From a study of it in the type locality, Forel concluded that its colonies contain no slaves. I have shown, however, that the female *aserva* establishes her colony with the aid of workers of *F. fusca* or its var. *subsericea* pillaged as pupae, but that the colony eventually becomes a pure *aserva* colony, because the workers of this subspecies fail to inherit their mother's predatory and dulotic instincts. This explains why Forel failed to find any *fusca* workers in the large colonies which he examined at Toronto. I have seen only two male specimens of *aserva*, both from South Harpswell, Maine, and one of these was immature.

7. F. SANGUINEA RUBICUNDA Emery.

F. sanguinea subsp. *rubicunda* Emery, Zool. jahrb. Syst., 1893, **7**, p. 647, pl. 22, fig. 2, ☿ ♀; Wheeler, Amer. nat., 1901, **35**, p. 711; Bull. Amer. mus. nat. hist., 1906, **22**, p. 74; Ants, 1910, p. 458, 570.

Worker. Length 5–7 mm.

Head shaped much as in the European *sanguinea*, with rather straight converging sides, feebly excised posterior border and prominent posterior angles; clypeal notch shallower and less pronounced; antennal scapes but slightly enlarged at their tips. Petiole broad, with thin, rather sharp superior border, usually notched in the middle.

Body, especially the gaster, somewhat more shining than in the typical *sanguinea*, owing to the pubescence being a little shorter and sparser.

Hairs, especially on the gaster, longer and more abundant, usually of a rich golden yellow color, but sometimes grayish or whitish. Hairs on the dorsal surface of the head, pro- and mesonotum numerous, and there are usually also a few erect hairs on the gula and petiolar border. Pubescence very distinct, fine, gray, short on the head and thorax, longer on the gaster. Femora with a row of hairs on their flexor surfaces; tibiae with short, appressed pubescence and a row of short bristles on their flexor surfaces.

Color of head, thorax, petiole, and appendages usually lighter and slightly more yellowish than in the European type; the head not darker than the thorax; mandibles but little darker than the head. Gaster black.

FEMALE. Length 7–9 mm.

Very similar to the worker in sculpture, pilosity, and color. Space between frontal carinae and sometimes also the clypeus infuscated; mesonotum usually immaculate; antennae and tibiae brownish; wings infuscated at the base, in some specimens more strongly than in the European type.

MALE. Length 7–9 mm.

Mandibles broad, dentate; clypeus carinate, convex, its anterior border feebly emarginate. Closely resembling the European type in color and pilosity, but the gaster is more shining, owing to its somewhat sparser pubescence. Petiole much thicker and with a blunt border, which is more faintly excised or sometimes even entire and transverse. Antennae black throughout, mandibles reddish only at their tips, which are dentate as in the type. Legs in mature specimens sordid yellow, with the femora more or less infuscated basally. Genitalia yellow, the appendages infuscated at their tips. Wings as in the female.

HOSTS (SLAVES). *F. fusca* var. *subsericea; F. cinerea* var *neocinerea; F. neogagates; F. pallidefulva schaufussi* and var. *fuscata.*

TYPE LOCALITY.— Pennsylvania (Emery).

New Jersey: Milltown (W. T. Davis); Delaware Water Gap (H. L. Viereck); Woodbury (Phila. Acad. Coll.); Newfoundland (Wheeler).

North Carolina: Black Mts. and Panther Gap, Blue Ridge (W. Beutenmüller).

Massachusetts: Ellisville, Woods Hole, Blue Hills (Wheeler); Springfield, Holyoke (G. B. King).

Connecticut: Colebrook (Wheeler).

Michigan: Marquette (M. Downing).

Illinois: Rockford (Wheeler).

Colorado: Prospect Lake, Colorado Springs (Wheeler).

Montana: Helena (W. M. Mann).

Ontario: Guelph (W. H. Wright).

This subspecies, which is not as common as the subspecies *subintegra* or even *subnuda,* varies considerably in different colonies in the color and character of the pilosity. Thus in my workers from the Black Mountains of North Carolina the hairs on the gaster are gray, very slender, and pointed, whereas in specimens from most other localities they are brilliant golden yellow. Emery cites a single worker from Labrador as belonging to *rubicunda*, but I believe that it was more probably a specimen of the subspecies *subnuda*. I prefer to cite Pennsylvania as the type locality because it falls within the range of *rubicunda* as indicated by my material and because Emery utilized in his description several worker and female specimens from that state.

I have shown that the female *rubicunda* founds her colony by kidnapping and rearing the pupae of *F. fusca* var. *subsericea*. After the colony is thus established the workers of *rubicunda* make periodical dulotic raids on colonies of *subsericea* and by rearing its pupae maintain a mixed colony. I have always taken *rubicunda* with *F. subsericea, neogagates* or *schaufussi* as slaves except at Colorado Springs, Colo., where the colonies on the shores of Prospect Lake contained instead *F. cinerea* var. *neocinerea*.

8. F. SANGUINEA RUBICUNDA var. SUBLUCIDA, var. nov.

WORKER. Length 5.5–6.5 mm.

Differing from the worker of the typical *rubicunda* in the more shining surface of the body, especially of the mandibles, frontal area, head, and gaster. The hairs and pubescence are well developed but grayish, and the pubescence is much sparser on the gaster. The head is proportionally larger, with more rounded sides and posterior corners, the clypeal notch is shallower and the thorax seems to be more slender; the petiole is narrower and has a blunter superior border, much like the petiole of the subspecies *subintegra*. The body is light red, with deep black gaster and brownish legs.

FEMALE (DEÄLATED). Length 8–9 mm.

Closely resembling the female of *rubidunda* but the thorax is proportionally smaller and narrower. The pubescence on the head, thorax, and gaster is longer than in the worker so that these parts appear to be less shining. One specimen has three fuscous spots on the mesonotum, the other has this region immaculate. The petiole is much like that of the worker and not so broad and sharp as in *rubicunda*.

HOST (SLAVE). *F. fusca* var. *subsericea*.

Described from two females and several workers taken from a single colony on the Stony Brook Reservation, near Boston, Mass. This form may deserve to rank as a distinct subspecies when more material is available. The frontal area is very smooth for a *sanguinea*, almost as smooth and shining as in the *rufa* forms. The thorax is rather slender and recalls the structure of this region in *F. munda* and *F. pergandei*.

9. F. SANGUINEA SUBNUDA Emery.

F. sanguinea rubicunda var. *subnuda* Emery, Zool. jahrb., Syst., 1895, **8**, p. 335, ☿.

F. sanguinea subsp. *subnuda* Wheeler, Ants, 1910, p. 458, 570.

Worker. Length 5–8 mm.

Head like that of the typical *rubicunda* but the clypeal emargination is much shallower, often reduced to a feeble sinuosity. Epinotum often more rounded and less angular in profile, especially in smaller workers, in larger ones, however, often as angular as in the typical *rubicunda* and *aserva*. Petiole rather broad, with sharp, entire, or very feebly sinuate superior border.

Surface like that of *rubicunda,* the gaster usually slightly more opaque.

Hairs grayish or yellowish, much less abundant than in the typical *rubicunda* and its var. *sublucida,* nearly always completely absent on the thoracic dorsum, gula, and petiolar border. There are only a few hairs on the upper surface of the head and those on the gaster are decidedly short and sparse. Pubescence on gaster dense but finer than on the typical *rubicunda,* concealing the surface; on the thorax and head very sparse or absent and often not perceptible under an ordinary magnification.

Color variable, but usually a light, rich red like that of the typical *rubicunda,* in some cases, however, more brownish; gaster black, as a rule, but occasionally with each segment brownish or reddish towards its base.

Female. Length 8–9 mm.

Closely resembling the worker in sculpture and pilosity, but the red portions of the body somewhat browner. Dark spots on the mesonotum faint or wanting. Wings colored as in the typical *rubicunda,* if anything somewhat more deeply. Clypeal border more deeply notched than in the worker.

Male. Length 8–9 mm.

Differing from the males of the preceding forms of *sanguinea* in having the anterior border of the clypeus entire and evenly rounded; its surface is convex and carinate. Mandibles dentate. Petiole somewhat more compressed anteroposteriorly and with a sharper border than in *rubicunda.* There are no erect hairs on the head and thorax and the hairs on the gaster are short and sparse. Pubescence short and dilute so that the surface of the head, thorax, and gaster is more shining than in the typical *rubicunda.*

Hosts (Slaves). *F. fusca* vars. *subsericea, argentea, subaenescens,* and *gelida;* large colonies often without slaves.

Type locality.— British Columbia: Yale, (Dieck).

British Columbia: Vancouver I.; Field, Carbonate, 2,800 ft., Lake

Minnewonka, Howser, Roger's Pass, Selkirk Mts. (J. C. Bradley); Golden (W. Wenman).

Alberta: Vermillion Pass (E. Whymper); Smith's Landing (H. V. Radford).

Saskatchewan: Methy Lake (R. Kennicott).

Manitoba: Winnipeg (S. H. Scudder).

Quebec: Mingan Island; Niapisca Island; Grand Grève, Gaspé (S. Henshaw); Kingsmere (Wheeler).

Ontario: Rat Portage (J. C. Bradley); Marshall's Bay near Arnprior (C. G. Hewitt).

Nova Scotia: Digby (J. Russell); Port Maitland (W. Reiff); Boisdale, Cape Breton I. (Amer. Mus. Coll.).

Newfoundland: Bay of Islands (Amer. Mus. Nat. Hist. Coll.).

Arizona: San Francisco Mts., 12,000 ft. (W. M. Mann).

New Mexico: Harvey's Ranch, Las Vegas Range, 9,600–10,000 ft. (Miss Ruth Reynolds and E. L. Hewett); Beulah, 8,000 ft. (T. D. A. Cockerell).

Colorado: Breckenridge (P. J. Schmitt); Ward, 9,000 ft., Pike's Peak, 10,000 ft. (T. D. A. Cockerell); Pike's Peak, 11,500 ft., Woodland Park, 8,500 ft., Ute Pass, 8,000 ft., Cheyenne Canyon, Manitou (Wheeler).

Montana: Helena (W. M. Mann).

Idaho: Troy (W. M. Mann).

Michigan: Isle Royale (O. McCreary).

Maine: South Harpswell (Wheeler).

Connecticut: Colebrook, 1,500 ft. (Wheeler).

The foregoing list of localities shows that *subnuda* is a boreal and alpine form like *aserva*, but unlike this subspecies confined very largely to the Rocky Mountains within the confines of the United States. As I have found no transitions between it and the typical *rubicunda*, I believe that it should rank as a subspecies and not as a variety. It is not always easy to separate it from *aserva*. Specimens from Golden and Howser, B. C., are very dark and much like *aserva*, except that the head, even in the smaller workers, is not darker in color than the thorax. The emargination of the clypeus is, however, extremely feeble in these specimens, even feebler than in *aserva* and *rubicunda*.

10. F. SANGUINEA SUBINTEGRA Emery.

F. sanguinea subsp. *rubicunda* var. *subintegra* Emery, Zool. jahrb. Syst., 1893, **7**, p. 648. ☿ ♀; Wheeler, Amer. nat., 1901, **35**, p. 713; Bull. Amer. mus. nat. hist., 1906, **22**, p. 84.

F. sanguinea subsp. *subintegra* Wheeler, Bull. Amer. mus. nat. hist., 1908, **24**, p. 627; Ants, 1910, p. 458, 570.

WORKER. Length 4–7 mm.

Head with the posterior corners and sides more rounded than in the preceding forms. Clypeus with broad but shallow emargination. Antennal scapes usually but slightly thickened towards their tips. Pro- and mesonotum not very convex, epinotum somewhat rounded in profile. Petiole thick anteroposteriorly, narrow seen from behind, convex in front, flattened behind, with blunt, usually entire superior border.

Surface of body rather smooth and somewhat lustrous or shining, especially the mandibles and posterior corners of the head; frontal area shining, except in the middle.

Pilosity and pubescence yellow, the former represented by a few hairs on the dorsal surface of the head, sometimes a few on the pronotum and by a number of scattered hairs on the gaster, longest at the tip and on the venter. Rarely there are a few hairs on the petiolar border, and on the gula. Pubescence abundant and dense on the gaster, somewhat finer on the remainder of the body but clearly visible under a lens magnifying 16 diameters. Surfaces of femora and tibiae with very fine, appressed pubescence, which is dense on their anterior and sparse on their posterior faces. Femora without hairs, tibiae with a row of graduated bristles on the flexor surface.

Red color of head, thorax, petiole and appendages usually tinged with yellow. Mandibles darker, with black teeth. Gaster brown.

FEMALE. Length 7–9 mm.

Closely resembling the worker in pilosity and color. Surface of body more opaque. Mandibles, clypeus, front, antennae, tibiae, and tarsi and sometimes also three spots on the mesonotum, brownish. Wings rather heavily infuscated at their bases. Petiole like that of the worker but broader.

MALE. Length 7–8 mm.

Mandibles with rather narrow blades, pointed, edentate. Clypeus with rounded, entire anterior border, carinate, often with an indistinct transverse impression just back of its anterior border and another near its posterior end. Petiole thick, transverse, with blunt, feebly and broadly excised dorsal margin.

Pubescence similar to that of the worker, more dilute on the gaster, so that this region appears more shining. Erect hairs very short, confined to top of head, mesonotum and scutellum.

Body black; antennae brown; legs and genitalia yellow; wings rather more heavily infuscated than in the female.

HOSTS (SLAVES). *F. fusca* and its vars. *subsericea, subaenescens, F. cinerea* var. *neocinerea, F. neogagates* and *vidua; F. pallidefulva schaufussi, nitidiventris, fuscata,* and *incerta.*

Type locality.— District of Columbia (Emery).

Newfoundland: Bay of Islands (L. P. Gratacap).

New Brunswick: St. Stephen (Cent. Exper. Farms Coll.).

Nova Scotia: Digby (J. Russell).

Quebec: Hull, Kingsmere (Wheeler).

Ontario: Guelph (W. H. Wright); Ottawa (Cent. Exper. Farms Coll.).

Maine: S. Harpswell, and Lower Goose Island (Wheeler).

Massachusetts: Sherborn (A. P. Morse); Woods Hole, Ellisville (Wheeler); Springfield (J. A. Allen); Essex County (G. B. King).

Connecticut: New Haven (H. L. Viereck): Colebrook (Wheeler).

New York: Bronxville, Mosholu (Wheeler); Staten Island (W. T. Davis).

New Jersey: Woodbury; New Brunswick (J. B. Smith); Lakehurst, Newfoundland (Wheeler).

Pennsylvania: Beatty (P. J. Schmitt).

Illinois: Rockford, Cherry Valley (Wheeler).

I believe that this form, too, should rank as a subspecies and not as a variety of *rubicunda*. Emery mentions workers from Beatty, Pa., which were transitional in the shape of the head and petiole between *rubicunda* and *subintegra*. I have seen similar specimens from a few of the localities recorded above, but such specimens in pilosity and in the brown color of the gaster are always easily referable to the latter subspecies. The smaller size, the peculiar color of the gaster, the more rounded shape of the head, the narrower, thicker, and blunter petiole of the worker, and the absence of mandibular teeth in the male sufficiently distinguished *subintegra* from *rubicunda*, but its separation from the next subspecies, *puberula* is not so easy. *F. subintegra* is the common form of *sanguinea* in the Eastern States and Canada at low elevations and in warm situations. I have shown that its queens establish their colonies in the same manner as the queens of *rubicunda*.

11. F. SANGUINEA SUBINTEGRA var. GILVESCENS, var. nov.

Worker. Length 4.5–5 mm.

Differing from the typical *subintegra* in the following characters:— The anterior border of the clypeus is so feebly notched as to appear merely somewhat truncated in the middle; the erect hairs are very short and sparse on the gaster, almost lacking on the thorax, sparse but somewhat longer on the head, absent on the gula. Color yellow, gaster, head, and antennae tinged with brownish, in more immature specimens the head and antennae are yellow and the gaster is only a little darker than the thorax.

Host (Slave). *F. fusca* var. *subsericea.*

Described from several specimens taken from a single colony at Tuckahoe, N. Y. To the same variety I refer a number of workers which I took from several nests at Calhoun, Waukesha County, Wisconsin, although these specimens are somewhat darker and more reddish. In these respects they are transitional to the typical *subintegra.*

12. F. sanguinea puberula Emery.

F. sanguinea subsp. *puberula* Emery, Zool. jahrb. Syst., 1893, **7**, p. 648, ☿; Wheeler, Ants, 1910, p. 458, 570.

Worker. Length 4–6 mm.

Head rather large, in large workers shaped like that of *rubicunda,* with less convex sides, and less rounded posterior angles than in *subintegra.* Clypeal notch broad and rather deep. Antennal scapes often distinctly thickened towards their tips. Thorax and petiole similar to those of *subintegra,* but the latter more compressed anteroposteriorly, less convex anteriorly and usually with a sharper upper border, which is sometimes feebly notched in the middle.

Surface of body as in *subintegra;* mandibles more shining because more densely and superficially striated and less distinctly punctate.

Hairs yellow, more abundant than in *subintegra,* present on the pro- and mesonotum, petiolar border and gula. Those on the gaster are long and slender. Anterior surfaces of tibiae with small, oblique hairs and without appressed pubescence. Pubescence grayish, moderately abundant on the gaster, finer and sparser but still visible on the head and thorax, long on the antennal scapes, especially towards their tips.

Color like that of *subintegra,* the head, thorax, and appendages being yellowish red, the gaster brown.

Female. Length 7–8 mm.

Closely resembling the worker in sculpture, pilosity, and color. Mandibles coarsely striatopunctate. Clypeal notch deep. Antennal scapes considerably enlarged towards their tips. Mesonotum without dark spots. Wings rather deeply infuscated at their bases.

Male. Length 7–8 mm.

Mandibles indistinctly toothed; clypeus convex, carinate, with feebly but distinctly emarginate anterior border. Petiole transverse, low and thick, with blunt, slightly excised superior border.

Pilosity and pubescence much as in the male of *subintegra,* the pubescence perhaps a trifle longer and more conspicuous on the legs.

Black; antennae dark brown; legs yellow, the femora sometimes infuscated. Genitalia brownish. Wings usually very deeply infuscated at their bases.

HOSTS (SLAVES). *F. fusca* vars. *argentea, subaenescens,* and *neoclara; F. cinerea* var. *neocinerea; F. pallidefulva nitidiventris*; *F. neogagates lasioides* var. *vidua.*

TYPE LOCALITY.— South Dakota: Hill City, (Emery).

Colorado: Manitou, Colorado Springs, Cheyenne Canyon, Ute Pass, Woodland Park (Wheeler); Breckenridge, West Cliff (P. J. Schmitt).

Utah: Stockton (T. Spalding).

Washington: Pullman (W. M. Mann); Olympia (T. Kincaid).

Montana: Helena (W. M. Mann).

New Mexico: Manzanares (Miss Mary Cooper); Alamogordo (G. v. Krockow); Gallinas Canyon (T. D. A. Cockerell).

Texas: Ft. Davis (Wheeler).

Missouri: Doniphan (P. J. Schmitt).

Illinois: Rockford (Wheeler).

This subspecies replaces *subintegra* at lower altitudes and in warmer situations in the Western States. Occasionally one finds specimens of the latter form which approach *puberula* in the somewhat longer pubescence on the legs and the more abundant hairs on the body. I have taken such specimens at Lakehurst, N. J. The males of *puberula* from Illinois are abnormally small (7 mm.), and in the shape of the clypeus resemble *subintegra.* Apart from the conspicuous differences in pilosity, the workers of the two forms can be separated in nearly all instances by the pubescence, which, on the anterior surfaces of the tibiae, is very fine, dense, and appressed in *subintegra,* but distinctly longer, sparser, coarser, and more oblique in *puberula,* so that in this form it takes on the appearance of minute hairs. I have seen no specimens of *puberula* which show this condition also on the antennal scapes; on these organs the fine, dense pubescence is merely a little longer but scarcely more oblique than in *subintegra.*

13. F. SANGUINEA OBTUSOPILOSA Emery.

F. sanguinea subsp. *obtusopilosa* Emery, Zool. jahrb. Syst., 1893, **7**, p. 648, ☿; Wheeler, Ants, 1910, p. 458, 570.

WORKER. Mandibles finely striated, feebly punctate. Clypeus rather deeply and broadly notched. Petiole narrow and thick, with blunt superior border, resembling the petiole of *F. pallidefulva.* Gaster opaque, with feeble metallic luster, its pubescence not dense but long and whitish. Erect hairs more abundant than in the other subspecies, whitish yellow, all nearly of the same length, enlarged

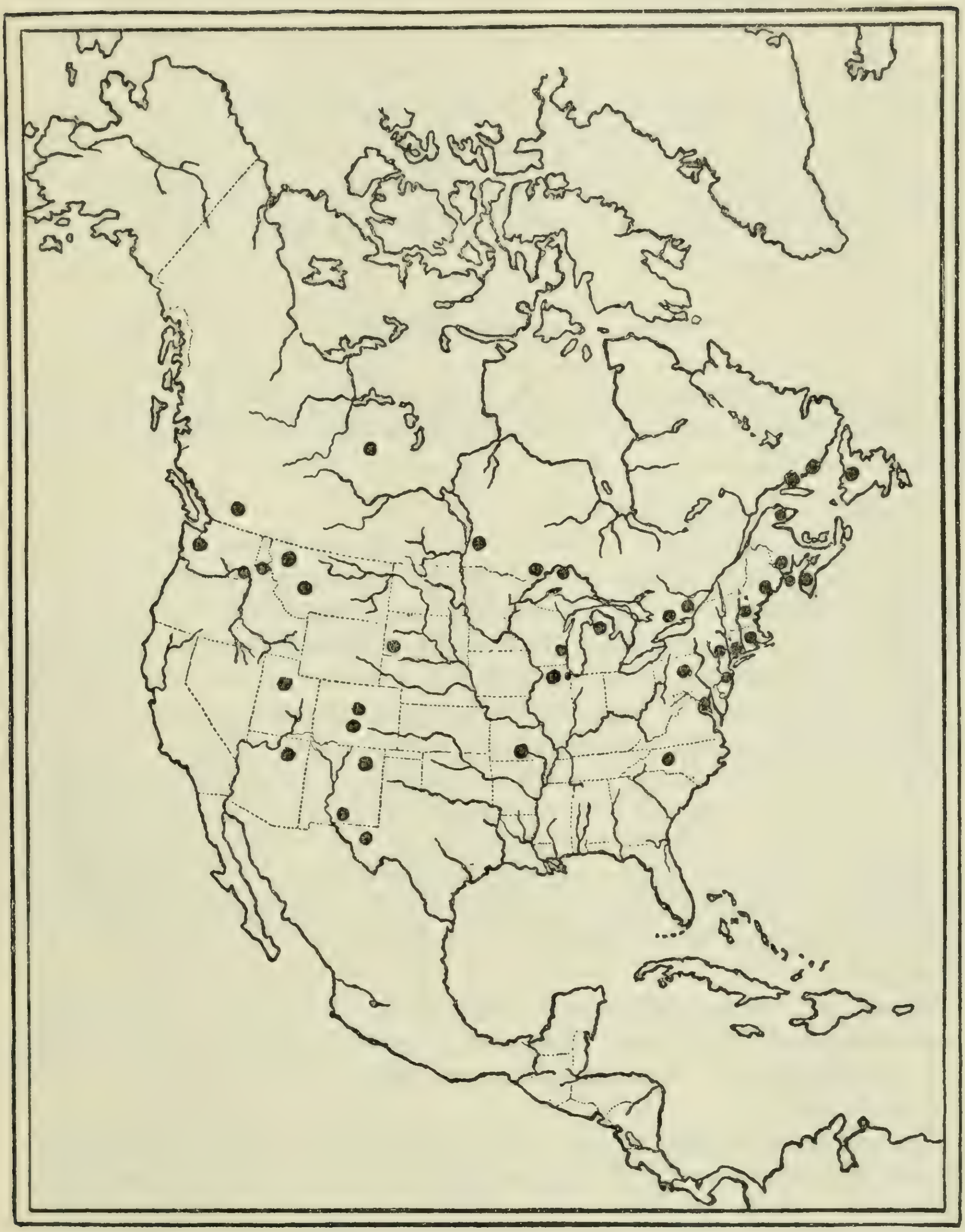

FIG. 1.— Distribution of the Nearctic forms of *Formica sanguinea.*

towards their tips which are truncate. The hairs of the thorax have the same form; and there are a few of them also on the border of the petiole (Emery).

Emery described this subspecies from a single worker taken in New Mexico. For some time I attributed several specimens in my collection from the same state to this subspecies, but closer examination shows them to belong to what I described as *F. munda.* I must admit, therefore, that I have never seen the true *obtusopilosa.* My reasons for believing that *F. munda* is a distinct species are given below.

14. F. MUNDA Wheeler.

F. pergandei var. Emery, Zool. jahrb. Syst., 1893, **7**, p. 647, ☿.

F. munda Wheeler, Bull. Amer. mus. nat. hist., 1905, **21**, p. 267, ☿ ♀; Ants, 1910, p. 458.

WORKER. Length 5–7 mm.

Mandibles 8-toothed. Head, excluding mandibles, usually somewhat longer than broad, with straight or slightly convex posterior border and long cheeks, converging anteriorly and slightly convex or flattened. Clypeus sharply carinate, with a rather deep and broad notch in its anterior border. Antennae slender, scapes not enlarged towards their tips. Thorax rather low and narrow, pro- and mesonotum not very convex, mesoëpinotal constriction shallow, epinotum long and low, its basal surface horizontal in profile and somewhat longer than the very sloping declivity into which it passes through a rounded angle. Petiole low and thick, convex in front, flattened behind, with a very obtuse, entire superior border. Seen from behind the border is transverse, broadly rounded, but passing rather abruptly into the straight sides, which converge below. Gaster small; legs slender.

Head and thorax subopaque, very finely shagreened. Mandibles, anterior portion of head, and especially the borders of the frontal area and sides of the clypeus, more shining. Mandibles sharply striatopunctate.

Pubescence grayish, sparse, except on the gaster where it is long and dense and conceals the shining surface, except at the intersegmental incisures. Hairs on the body rather abundant, glistening white, obtuse, suberect, and rather long on the upper surface of the head, thorax, and gaster; on the gaster very regularly distributed. Petiolar border with a row of similar hairs. Legs invested with small, sparse, appressed hairs; femora and tibiae with a row of erect or oblique hairs on their flexor surfaces.

Head, thorax, and antennae red; petiole and gaster black, the former often with a reddish tinge. Mandibular teeth black. Lower pleurae and in many specimens also the vertex of the head, infuscated. Legs red; coxae, femora, and tibiae more or less infuscated, except at the articulations.

Female. Length 7.5–8 mm.

Head small, narrower than the thorax; antennal scapes extending nearly $\frac{1}{3}$ their length beyond the posterior corners of the head. Resembling the worker in pilosity, sculpture, and coloration, except in the following characters: — The hairs are of a yellowish cast, and on the gaster are pointed and of the same thickness as on the head and thorax, although they are long and in certain lights conspicuous, especially towards the tip of the body. Pleurae clouded with fuscous; posterior portion of head, posterior edge of pronotum, and anteromedian and two parapsidal blotches on the mesonotum, fuscous. Metanotum and scutellum, except its anterior border, black. Petiole varying from dark red to blackish, of the same shape as in the worker, except that in profile its superior border is much sharper in some specimens. Wings whitish hyaline, with pale brown veins and stigma.

Type locality.— Colorado: Canyon City (P. J. Schmitt).

Colorado: Breckenridge, West Cliff (P. J. Schmitt); Colorado Springs, Salida, Boulder, Wild Horse (Wheeler); South Boulder Canyon (T. D. A. Cockerell); Troublesome (S. A. Rohwer).

New Mexico: Glorieta, Old Pecos Pueblo (T. D. A. Cockerell).

South Dakota: Medicine Root, Pine Ridge Ind. Reserv. (Thompson). Harding County (S. S. Visher).

Montana: Helena (W. M. Mann).

Alberta: Medicine Hat (J. C. Bradley).

The worker of this species differs from *sanguinea* and resembles *F. pergandei* in the structure of the thorax. The head, especially of large workers, is more like that of small *sanguinea* workers and broader than in *pergandei*. From this latter species and from all the subspecies of *sanguinea*, except, perhaps, *obtusopilosa*, *munda* differs in the peculiar thick, blunt hairs, especially on the gaster. The female is readily distinguished from the female *sanguinea* by the smaller head and longer antennal scapes. Some years ago Professor Emery informed me (*in litteris*) that the specimens which I later described as *F. munda* were identical with the ones he regarded in his "Beiträge" as representing a variety of *pergandei* from Colorado. I infer therefore that *F. munda* cannot be a synonym of his *F. sanguinea obtusopilosa,* as one might be led to believe from a study of his brief description of that subspecies.

F. munda lives in grassy places, especially in irrigated plains and pastures at altitudes of about 6,000–7,000 ft. The colonies, which are rather small and comprise only a few hundred workers, make small obscure crater nests like those of *F. schaufussi* and its varieties in the Eastern States. I have never found *munda* nesting under stones, and in no colony have I been able to find any slaves. There is, indeed, absolutely nothing to indicate that this ant is ever parasitic or dulotic.

15. F. PERGANDEI Emery.

F. pergandei Emery, Zool. jahrb. Syst., 1893, **7**, p. 646, pl. 22, fig. 1, ☿; Wheeler, Bull. Amer. mus. nat. hist. 1905, **21**, p. 268; Ants, 1910, p. 458, 470.

WORKER. Length 5.5–6.5 mm.

Mandibles 8-toothed. Maxillary palpi short and very slender. Head longer than broad, with long, flat or slightly concave cheeks, converging anteriorly; posterior border straight. Clypeus carinate, not very convex, its anterior margin impressed and rather broadly and deeply notched in the middle. Antennae slender, the scapes not distinctly enlarged towards their tips. Thorax rather long and slender, pro- and mesonotum not very convex, mesoëpinotal constriction well developed, epinotum in profile roundly angular, with subequal base and declivity, the former horizontal, the latter sloping. Petiole narrow, more convex anteriorly than posteriorly, with an obtuse, entire superior border.

Mandibles and clypeus shining, the former finely striated and indistinctly and sparsely punctate, the latter indistinctly, longitudinally rugulose. Head, thorax, and petiole smooth, subopaque, gaster and legs shining. Frontal area shining, except in the center.

Hairs slender and grayish, very sparse on the pronotum and dorsal surface of the head, more abundant on the gaster. There are a few erect hairs on the gula, at least in some specimens. Pubescence very sparse on the head and thorax, longer on the gaster, but not sufficiently dense to conceal its shining surface. Legs and scapes with minute subappressed hairs; tibiae with a row of slanting bristles on their flexor surfaces.

Brownish testaceous; mandibles darker; mandibular teeth and gaster black.

HOST (SLAVE?). *F. pallidefulva.*

TYPE LOCALITY.— District of Columbia: Washington (Th. Pergande). Massachusetts (J. G. Jack).

This species seems to be extremely rare. I have seen two cotypes kindly sent me by Prof. Emery and two specimens from Massachusetts which agree with these. From the fact that Pergande found the species living with *F. pallidefulva,* Emery has inferred that it is dulotic like *sanguinea.*

F. pergandei is readily distinguished from *sanguinea* by its narrow head and body, its brown color and smoother surface. From *F. munda* it differs in having the erect hairs slender and pointed, in its duller coloration, narrower head and shorter maxillary palpi.

16. F. EMERYI, sp. nov.

F. pergandei Wheeler, Bull. Amer. mus. nat. hist., 1905, **21**, p. 268.

WORKER. Length 4.5–6 mm.

Head a little longer than broad, a little narrower in front than behind, with straight cheeks and posterior border. Mandibles 8-toothed. Maxillary palpi short. Clypeus sharply carinate, its anterior margin neither produced nor impressed but feebly and narrowly notched in the middle. Antennae rather robust; scapes slightly enlarged towards their tips; funicular joints subequal; joints 2–5 a little more slender than the succeeding joints. Thorax rather long, pro- and mesonotum depressed, mesoëpinotal constriction narrow, the posterior surface of the mesonotum falling suddenly to the level of the metanotum; epinotum angular, with subequal base and declivity, the former with a very faint transverse impression in the middle. Petiole narrow; cuneate in profile, with straight posterior and very feebly convex anterior surface, its border entire, rounded and rather sharp. Gaster elliptical, more elongate than in any of the preceding species. Legs stout and rather long.

Mandibles shining, very finely and rather superficially striated, with very fine, scattered punctures. Remainder of body opaque, its surface very finely and uniformly shagreened; gaster with a slightly metallic luster.

Pilosity and pubescence gray, the former represented by only a few erect hairs on the front and clypeus and two transverse rows of sparse hairs on each gastric segment, which are somewhat longer towards the tip and on the venter. Tibiae each with a row of bristles on their flexor surfaces. Pubescence extremely short and inconspicuous on the head, thorax, petiole, and legs, a little longer and much denser on the gaster, so that this region has a grayish tint.

Brown; gaster black; mandibles red; dorsal portion of head infuscated or blackened.

FEMALE. Length 7–7.5 mm.

Closely resembling the worker in sculpture, pilosity, and color. Mandibles much more coarsely striatopunctate. Clypeus with broader but very shallow emargination. Head large, broader than the thorax, scarcely longer than broad. Antennal scapes reaching only a distance equal to their own diameter beyond the posterior corners of the head. Infuscation of top of head deeper and more extensive than in the worker, covering also the cheeks and clypeus. Mandibles brown. Mesonotum immaculate. Metanotum and posterior border of pronotum infuscated. Petiole like that of the worker in shape. Wings whitish hyaline, without any trace of infuscation, veins and stigma brown.

Host (Slave). *F. neogagates.*

Described from nine workers and four females taken Aug. 8, 1903, from a small colony in the open fields at Broadmoor, near Colorado Springs, Colo. The nest contained several small workers of *F. neogagates,* which were in all probability the slaves of the new species, since winged females of the latter were found in the nest. At first sight *F. emeryi* appears to be merely a variety or subspecies of *pergandei,* but closer examination shows many dissimilarities, especially the smaller size, the greater breadth of the head, the much feebler pilosity, the deeper color, the more opaque surface, and the shape of the thorax in profile. The slight transverse depression in the base of the mesonotum is constant in all my specimens. The female may be readily distinguished from the females of *sanguinea* and *munda* by its color, from *sanguinea* also by the pale, colorless wings, and from *munda* by its much larger head and shorter antennal scapes.

17. F. MANNI, sp. nov.

Worker. Length 3.5–4.5 mm.

Body slender. Head, excluding the mandibles, longer than broad, a little narrower in front than behind, with straight sides and feebly convex posterior border. Clypeus carinate, its anterior border feebly and rather broadly notched in the middle. Frontal carinae subparallel behind. Antennae slender, scapes not incrassated toward their tips. Thorax long, pro- and mesonotum moderately convex; mesoëpinotal constriction shallow; epinotum angular in profile, with subequal base and declivity. Petiole rather narrow; in profile cuneate, rather thick at the base, gradually narrowed towards the summit, with nearly flat anterior and posterior surfaces, the border rather sharp; seen from behind entire or very feebly excised in the middle. Legs rather long.

Body very finely shagreened, shining, especially the gaster; the clypeus and mandibles somewhat more opaque, finely striated, the former also sparsely punctate.

Hairs whitish, long, rather slender, erect, sparse; conspicuous on the upper surface of the head, clypeus, gula, thoracic dorsum, petiolar border, gaster, and fore coxae. Pubescence very short and sparse, most clearly visible on the gaster and legs but far from concealing the ground surface; scarcely perceptible on the cheeks and pleurae.

Rich red, legs a little paler and more yellowish; small workers darker and more brownish; tips of antennal funiculi and sometimes also the posterodorsal portion of the head in the large workers slightly infuscated; gaster always deep black throughout.

FEMALE (DEÄLATED). Length 6–7 mm.

Closely resembling the worker in sculpture, pilosity, and color. The notch in the clypeus is very broad and shallow and the carina very blunt or lacking. The petiole is broad, with a flat, very sharp border. The mesonotum bears three faint brownish blotches, the wing-insertions and sutures of the thorax are blackish and the base of the first gastric segment is red, the posterior borders of the segments yellowish.

TYPE LOCALITY.— Washington: Kiona, (W. M. Mann).

Washington: Wapata, Wenatchee, Ellensburg (W. M. Mann).

California: Owen's Lake (H. F. Wickham).

The series of specimens includes many workers and three females, two from Kiona and one from Owen's Lake. At first sight this species, on account of its smooth and shining body and the character of the pubescence, appears to belong in the *fusca* group, but the structure of the clypeus seems to associate it more naturally with *sanguinea.* In the shape of the body it shows an even closer relationship to *F. pergandei, munda,* and *emeryi.* The small size of the female seems to indicate that it is a parasitic species. Mr. Mann informs me that the colonies are small and nest under stones in dry, hot, and often sandy, desert country.

18. F. PERPILOSA Wheeler.

F. fusca subpolita var. *perpilosa* Wheeler, Mem. revist. Soc. cient. Ant. Alzate, 1902, **17**, p. 141; Herrera, Boll. Comision parasit. agric., 1902, **1**, p. 404.

WORKER. Length 3–5.5 mm.

Head in large workers, excluding the mandibles, about as broad as long, a little narrower in front than behind, with straight lateral and posterior borders. Clypeus carinate, its anterior border rounded, entire, or in some specimens slightly truncated or even feebly emarginate in the middle. Antennae rather stout; scapes somewhat thickened towards their tips; basal joints of funiculus narrower but not longer than the penultimate joints. Frontal carinae diverging behind. Eyes rather small. Maxillary palpi moderately long. Pro- and mesonotum, especially the latter, convex; mesoëpinotal constriction short and rather deep; epinotum in profile with subequal base and declivity, both straight and forming a large, obtuse angle with each other. Petiole narrow, cuneate in profile, thick at the base, its anterior surface rather strongly convex, its posterior surface flat, its border obtuse, seen from behind rounded and entire. Legs rather stout.

Body and legs shining; very delicately shagreened, more coarsely on the metapleurae. Mandibles and clypeus subopaque, very finely and densely longitudinally striated, the former also with small, sparse, shallow punctures. Frontal area smooth and shining.

Head, throax, border of petiole, gaster, and fore coxae beset with long, erect, subobtuse, rather slender, silvery white hairs; those on the gula being as long as those on the upper surface of the head. Legs with only a row of hairs on the flexor surfaces of the femora and the usual row of bristles on the corresponding surfaces of the tibiae. Pubescence white, long, and sparse on the gaster, shorter and even sparser on the head, thorax, and legs, very fine and dense on the scapes.

Yellowish red; gaster black; in small workers the posterodorsal portion of the head, and the upper surface of the thorax and petiolar border often dark red or brownish.

FEMALE. Length 7.5–9 mm.

Closely resemblng the worker in color, sculpture, and pilosity, but the posterodorsal portion of the head, three large blotches on the mesonotum, the metanotum, and often also the posterior portion of the scutellum, and portions of the meso- and metapleurae fuscous. Gaster sometimes dark reddish brown, with a pale red spot at the base of the first segment. Wings colorless, with brown veins and black stigma. The middle of the clypeal border is flattened, and has a broad but shallow, sinuous excision. The petiole is broad, with a compressed, sharp border which is often produced upward in the middle as a blunt angle.

MALE. Length: 7–8 mm.

Mandibles rather short and broad, pointed, edentate. Head broad behind, with straight border, much narrowed in front, with straight cheeks. Eyes large. Clypeus convex, with entire, broadly rounded anterior border. Thorax and gaster rather slender. Petiole low and transverse, somewhat compressed anteroposteriorly, especially above, so that the border is less blunt than in the males of many other species of Formica; seen from behind the border is feebly and broadly excised. Stipes of genitalia with their tips projecting some distance beyond the volsellae and sagittae.

Head and thorax, including the frontal area, opaque. Mandibles, pleurae, and gaster somewhat shining.

Hairs and pubescence grayish, both very abundant, covering the head, thorax, and gaster; the hairs erect and rather short, the pubescence very long; eyes, scapes, and legs hairless.

Black; genitalia heavily infuscated; mandibles brown with yellowish tips; legs yellow, terminal tarsal joint of each foot black; wings uniformly gray, or smoky, with brown veins and black stigma.

Type locality.— Colorado: Canyon City (P. J. Schmitt).

Colorado: Cotopaxi (P. J. Schmitt).

New Mexico: Paraje, Las Valles (T. D. A. Cockerell); Alamogordo (G. v. Krockow).

Arizona: Tucson, Benson (Wheeler); Tempe (T. D. A. Cockerell).

Nevada: Las Vegas (J. C. Bradley).

Texas: San Esteban near Marfa, Langtry, Ft. Davis (Wheeler); Eagle Pass (J. D. Mitchell).

Mexico: Coahuila (A. F. Rangel).

This ant is certainly not a form of *subpolita,* nor does it belong with *fusca,* as I formerly supposed. It is closely related to the preceding species (*F. manni*), but differs in the greater size of the worker and especially of the female, the more robust body and antennae, more convex mesonotum, more abundant and longer pilosity and pubescence. *F. manni* might, perhaps, be regarded as a subspecies of *perpilosa.* The notch in the clypeus of the worker of the latter species is shallower and less constant, especially in small individuals than in *manni.*

Since the original account of this species was published ten years ago, I have had several opportunities of studying it in Arizona and Western Texas. It is preëminently a species peculiar to irrigated lands and river bottoms in the deserts of the southwest. There it nests in rather populous colonies about the roots of bushes or trees, often forming obscure craters or low mound nests, not unlike the nests of *F. subsericea* in the Eastern States. I have never found it nesting under stones. It is a very active and aggressive ant, and, as Herrera has shown, is of some little economic value as a boll-weevil exterminator. There is not the slightest indication that it is either a temporary social parasite or a slave-holder.

19. F. BRADLEYI, sp. nov.

Worker. Length 3.5–5 mm.

Head, excluding the mandibles, a little longer than broad, a little narrower in front than behind, with straight sides and straight or feebly convex posterior border. Eyes rather large. Clypeus convex, carinate, its anterior border not produced, broadly rounded, with a very shallow, broad excision in the middle. Frontal carinae subparallel behind. Antennae rather stout, the scapes slightly thickened towards their tips; second to fourth funicular joints somewhat more slender but scarcely longer than the antepenultimate joints. Maxillary palpi moderately long. Pro- and mesonotum, especially the latter, rather convex; mesoëpinotal constriction rather deep; epinotum in profile with subequal base and declivity, both straight and forming a large obtuse angle with each other. Petiole rather narrow, thick at the base with convex anterior and flat posterior surface and very blunt border, which is rounded and entire when seen from behind. Gaster rather large. Legs moderately stout.

Surface of body distinctly shagreened, shining; clypeus and mandibles densely longitudinally striate, the latter subopaque and also sparsely punctate. Frontal area very smooth and shining.

Hairs short, stout, obtuse, pale yellow, abundant, and erect, covering the dorsal and gular surfaces of the head, the thorax, fore coxae, petiole, and gaster; absent on the cheeks and pleurae. Pubescence rather sparse on the gaster, but slightly dimming the shining surface, shorter and less conspicuous on the head and thorax. Femora and tibiae with a row of hairs on the flexor surfaces; tibiae also with a few short subappressed hairs near the base on the extensor surface.

Light ferruginous red; mandibles a little darker; gaster if anything a little paler and more yellowish than the thorax.

MALE. Length 7 mm.

Mandibles broad, edentate. Head, excluding the mandibles, as broad as long, with convex, broadly rounded posterior border and much rounded posterior corners, short cheeks and very large, convex eyes. Clypeus sharply carinate, with entire, broadly rounded anterior border. Thorax and gaster slender. Petiole low and transverse, very thick and very bluntly rounded above; seen from behind its summit is slightly impressed in the middle. Genitalia with the tips of the stipes projecting beyond the volsellae and sagittae.

Surface of body, including the head and thorax as well as the gaster, shining; mandibles and clypeus more opaque; frontal area smooth and shining.

Hairs and pubescence grayish, more abundant than in the worker, the hairs of the same length, but the pubescence longer. Eyes and scapes hairless; legs with only a row of erect hairs on the flexor surfaces of the tibiae and femora. Pubescence on the legs much shorter than on the body.

Black; genitalia fuscous; tips of mandibles and legs beyond the tips of the coxae, yellow; last tarsal joint on each foot and basal portion of femora blackish. Wings grayish hyaline, the veins and stigma both of the same brown tint.

TYPE LOCALITY.— Colorado: Georgetown, (P. J. Schmitt). Alberta: Medicine Hat (J. C. Bradley).

Described from three workers and two males. A large number of the workers from Alberta are somewhat less shining but agree in other respects with the types.

The worker of this species is easily distinguished from all the other forms in the *sanguinea* group by its uniform red color and dense pilosity, which is much like that of *cinerea*. Indeed, were it not for the emarginaton of the clypeal border, it might be placed in the *fusca* group. The male is also very peculiar in its shining head and thorax, the unusual shape of the head, large size of the eyes and dense pilosity.

Rufa Group.

20. Formica rufa rufa Linné.

F. rufa Linné, Syst. nat., ed. 10, 1758, **1**, p. 580; de Geer, Mem. hist. ins., 1771, **2**, p. 1053, pl. 41, 42, fig. 1–11; Fabricius, Syst. ent., 1775, p. 391; Spec. ins., 1781, **1**, p. 489; Mant. ins., 1787, **1**, p. 308; Latreille, Essai hist. fourmis France, 1798, p. 39, ☿ ♀ ♂; Hist. nat. fourmis, 1802, p. 143, pl. 5, fig. 28, a, b, g, h.; Fabricius, Syst. Piez., 1804, p. 398, ☿ ♀ ♂; Latreille, Hist. nat. ins., 1805, **13**, p. 255; Gen. Crust. ins., 1809, **4**, p. 126; F. Smith, Trans. Ent. soc. Lond., 1842, **3**, p. 151–154; Nylander, Act. Soc. sci. Fennica, 1846, **2**, p. 902, pl. 18, fig. 16, ☿ ♀ ♂; Förster, Hymen. stud., 1850, **1**, p. 13, ☿ ♀ ♂; F. Smith, Trans. Ent. soc. Lond., 1855, ser. 2, **3**, p. 100, pl. 9, fig. 13, ☿ ♀ ♂; Nylander, Ann. sci. nat. Zool., 1856, ser. 4, **5**, p. 60, pl. 3, fig. 3; Mayr, Progr. realsch. Pest., 1856, p. 9, ☿ ; F. Smith, List Brit. anim. Brit. mus., 1858, pt. 6, p. 3; Mayr, Europ. Formicid. 1861, p. 46, 48, ☿ ♀ ♂; Forel, Denks. Schweiz. gesell. naturw., 1874, **26**, p. 52, 55, 57, 364, ☿ ♀ ♂; Bull. Soc. Vaud. sci. nat., 1875, ser. 2, **14**, p. 57, 59; Ern. André, Spec. Hymén. Europe, 1882, **2**, pt. 14, p. 184, 187, 189, pl. 1, 5, 6, 9, ☿ ♀ ♂; Lubbock, Ants, bees, wasps, ed. 5, 1882, p. 441, pl. 2, fig. 5, ☿ ; Dalla Torre, Catalog. Hymen., 1893, **7**, p. 206–209; Ruzsky, Formicar. Imper. Ross., 1905, p. 320, fig. 59–62; Emery, Deutschr. ent. zeitschr., 1909, p. 184.

F. dorsata, v. d. Hooven, Bijdr. natuurk. vet., 1826, **1**, p. 441.

F. obsoleta Zetterstedt, Insect. Lappon., 1838, **1**, p. 449, ☿ ♀.

F. lugubris Zetterstedt, Insect. Lappon., 1838, **1**, p. 449, ♂.

F. polyctena Förster, Hymen. stud., 1850, **1**, p. 15, ☿ ♀ ♂; Schenck, Jahrb. Ver. nat. Nassau, 1852, **8**, p. 25, 137, ☿ ♀ ♂; Stettin. ent. zeit., 1853, **14**, p. 160.

F. truncicola Förster, Hymen. stud., 1850, **1**, p. 21 ☿ , (excl. ♀ ♂).

F. piniphila Schenck, Jahrb. Ver. nat. Nassau, 1852, **8**, p. 28, 138, ☿ ♀ ♂.

F. apicalis F. Smith, List Brit. anim. Brit. mus., 1858, pt. 6, p. 49, ♀.

WORKER. Length 4–9 mm.

Resembling *F. sanguinea.* Body rather robust. Head subrectangular, about as long as broad, a little narrower in front than behind, with straight posterior and very feebly convex lateral borders. Mandibles 8-toothed. Clypeus strongly carinate, with entire anterior border. Antennae rather stout, penultimate much thicker and shorter than the basal funicular joints. Pro- and mesothorax very convex, hemispherical. Mesoëpinotal constriction pronounced; epinotum shaped much as in *sanguinea,* but more rounded and less angular. Petiole broad, compressed anteroposteriorly, with a sharp border, which is entire or feebly emarginate in the middle. Gaster large, broadly elliptical, legs long and robust.

Body opaque; mandibles shining, finely striatopunctate; frontal area glabrous and shining. Gaster glossy or feebly shining.

Pubescence fine and abundant; suberect hairs short, usually very sparse on the head and thorax, more abundant on the gaster. Gula almost always with a few erect hairs; eyes hairy. Tibiae with only a few minute slanting hairs in addition to the row of bristles on the flexor surfaces.

Dark or pale red; front, vertex and antennae dark brown to blackish brown; clypeus sometimes with a median longitudinal brown streak; pronotum usually with a small brown or black spot not reaching the posterior border of the segment; gaster blackish brown, its base somewhat reddish.

FEMALE. Length 9–11 mm.

Similar to the worker. Head and thorax opaque, but gaster very smooth and shining. Erect hairs short and sparse, absent on the gaster. Pubescence fine, most distinct on the legs and scapes. Red; front, vertex, mesonotum, the anal region, spots on the pleurae, tibiae, tarsi, tips of femora, middle of clypeus, and gaster, with the exception of its base, dark brown or black. Mandibles dark red, subopaque, coarsely striatopunctate. Wings slightly infuscated, with light brown veins and stigma.

MALE. Length 9–11 mm.

Body stout. Head rather small, mandibles edentate or very rarely dentate; masticatory border sharp, with apical point. Clypeus carinate, convex, with entire, angular anterior border. Petiole thick, its anterior and posterior surfaces flattened and its superior border very blunt, rounded and feebly excised in the middle.

Body, including the frontal area, opaque, upper surface of gaster shining.

Erect hairs black and dense on the head and thorax, sparse on the eyes and petiole, almost absent on the gaster.

Black; genitalia, and legs, except the bases of the femora, yellowish or brownish red. Wings like those of the female.

HOST (TEMPORARY). *F. fusca.*

North and Middle Europe, south as far as the Pyrenees and southern slopes of the Alps; Caucasus, Siberia; occurring only in the mountains in Southern Europe.

The typical *F. rufa* constructs large mound-nests of vegetable débris, usually pine-needles, in open forests, preferably of coniferous trees. A single colony may have several of these nests, which are connected with one another by run-ways. New colonies (not new nests!) are formed, as Wasmann and I have shown, by temporary social parasitism, the recently fecundated female finding a home in a *F. fusca* colony and permitting these ants to bring up her young. The *fusca* queen is either destroyed by the intrusive *rufa* queen or by her own offspring, so that when the *fusca* workers eventually die off, a pure colony of *rufa* remains. New nests are formed by adoption of *rufa* queens which leave the parental formicary with detachments of workers.

The forms *F. polyctena* Förster and *F. piniphila* Schenck are based on specimens which differ somewhat from the typical form in pilosity; *polyctena* having the head and thorax almost hairless, whereas *piniphila* is more pilose.

21. F. RUFA RUFA var. MERIDIONALIS Nassonov.

F. rufa var. *meridionalis* Nassonov, Arb. Lab. zool. Univ. Moskau, 1889, **4**, p. 17, ☿ ; Ruzsky, Formicar. Imper. Ross., 1905, p. 330; Emery, Deutsch. ent. zeitschr., 1909, p. 186.

WORKER. Differing from the typical form in color, the red parts being brownish yellow, the legs brown. Hairs very sparse.

Siberia.

It is not impossible, as Emery seems to imply, that this variety may be based on immature specimens of the typical *pratensis*.

22. F. RUFA RUFA var. RUFOPRATENSIS Forel.

F. rufa var. *rufopratensis* Forel, Denks. Schweiz. gesell. naturw., 1874, **26**, p. 53, ☿ ♀ ♂; Emery, Deutsch. ent. zeitschr., 1909, p. 186.

WORKER and MALE transitional in color and pilosity, and FEMALE in the smoothness of the gaster, between the typical *rufa* and the subspecies *pratensis*. These various characters are combined in the most manifold manner and degrees in different specimens.

North and Middle Europe.

According to Forel, this variety is usually smaller than the typical *rufa* and *pratensis*, more like the former in pilosity and more like the latter in color, but the reverse conditions are also found. The formicaries, too, are intermediate in all respects.

23. FORMICA RUFA RUFA var. SANTSCHII, nom. nov.

F. rufa var. *alpina* Santschi, Bull. Soc. ent. France, 1911, p. 349, 1 fig., ☿; Forel, Rev. Suisse zool., 1911, **19**, p. 457; Emery, Deutschr. ent. zeitschr., 1912, p. 672.

WORKER. Differing from the typical *rufa* in having the head proportionally longer and narrower (one fourth longer than broad), the scapes longer and extending further beyond the posterior corners of the head, the thorax narrower, the pro- and mesonotum more convex and the gaster a little larger. In size, color, and pilosity like the typical form.

Mountains north of Sondrio, Northern Italy (G. Valerio). Also in Great Britain, according to Donisthorpe, and Norway, according to Forel.

Santschi believes that this variety of *rufa* may prove to be merely an abnormality produced by entoparasitism of Mermis or Pelodera or by ectoparasitism of other insects. The name *alpina* is preoccupied for a variety of *F. adamsi* Wheeler.

24. F. RUFA PRATENSIS Retzius.

F. pratensis Retzius, Gen. & spec. ins., 1783, p. 75, ☿; Roger, Verz. Formic., 1863, p. 13; Ern. André, Rev. mag. zool., 1874, ser. 3, **2**, p. 184; Forel, Bull. Soc. Vaud. sci. nat., 1875, ser. 2, **14**, pt. 75, p. 58, 61; Ern. André, Spec. Hymén. Europe, 1882, **2**, pt. 14, p. 184, 189, ☿ ♀ ♂; Mayr, Fedtschenko's Turkestan. Formicid., 1877, p. 6; Lubbock, Ants, bees, wasps, ed. 5, 1882, p. 441; Forel, Ann. Soc. ent. Belg., 1886, **30**, p. 136, 138; Wasmann, Deutsch. ent. zeitschr., 1887, **31**, p. 109; Forel, Ann. Mus. St. Petersbourg, 1904, **8**, p. 385.

F. rufa Christ, Naturg. ins., 1781, p. 510, pl. 60, f. 7, ♀ ♂; Huber, Recherches moeurs fourm. indig., 1810, p. 320, ☿ ♀ ♂.

F. congerans Nylander, Acta Soc. Fennica, 1846, **2**, p. 906, ☿; Ibid., 1849, **3**, p. 26, 30, ♂; Förster, Hymen. stud., 1850, **1**, p. 17, ☿ ♀ ♂; Mayr, Verh. Zool. bot. ver. Wien, 1855, **5**, p. 332, ☿ ♀ ♂; Europ. Formicid., 1861, p. 46–48, ☿ ♀ ♂; F. Smith, List Brit. anim. Brit. mus., 1858, pt. 6, p. 2, pl. 3, f. 1, 7–9.

F. rufa st. *pratensis* Forel, Denks. Schweiz. gesell. naturw., 1874, **26**, p. 52, ☿ ♀ ♂.

F. pratensis Dalla Torre, Catalog. Hymen., 1893, **7**, p. 204.

F. rufa subsp. *pratensis* Ruzsky, Formicar. Imper. Ross., 1905, p. 337; Emery, Deutsch. ent. zeitschr., 1909, p. 186, ☿ ♀ ♂.

WORKER. Whole body and appendages, except antennae, covered with rather short but dense, suberect hairs. Eyes hairy. Fundamental colors as in the typical *rufa,* but the black spot on the head is larger, the spot on the pronotum reaches the posterior border of the segment and there fuses with a black spot on the mesonotum. Legs very largely brown; gaster entirely black.

FEMALE. Very similar to the female of the typical *rufa;* hairs sparse; eye sparsely hairy. Gaster subopaque or opaque, pubescent, brownish black except for a small basal and anal red spot.

MALE. Usually, but not always distinguishable from the male of *rufa* merely by its more abundant pilosity.

HOST (TEMFORARY). *F. fusca.*

Northern and Middle Europe, Siberia, Island of Sakhalin; in Europe ranging southward to Southern Italy in the high mountains.

This subspecies prefers to nest in meadows, fields, or along the borders of woods and hedges. The nests are similar to those of *rufa* and are single or in groups, but of average smaller size.

F. rufa pratensis is, like *rufa,* a temporary social parasite on *F. fusca.* In Southern Europe it probably prefers the var. *glebaria* of this ant as a host.

25. F. RUFA PRATENSIS var. NIGRICANS Emery.

F rufa var. *nigricans* Emery, Deutsch. ent. zeitschr., 1909, p. 187, ☿ .

WORKER. Color darker than that of the typical *pratensis,* the black spots on the thorax more extensive; legs brownish black with brownish red coxae and knees.

This is a southern form occurring in the Maritime Alps, Spain, and the Apennines (Vallombrosa), according to Emery.

26. F. RUFA PRATENSIS var. TRUNCICOLO-PRATENSIS Forel.

F. rufa st. *pratensis* var. *truncicolo-pratensis* Forel, Fourmis Suisse, 1874, p. 53; Ruzsky, Formicar. Imper. Ross., 1905, p. 348, ☿ ♀ ♂.

F. rufa pratensis var. *truncicolo-pratensis* Emery, Deutsch. ent. zeitschr., 1909, p. 187.

WORKER, FEMALE, and MALE in color and pilosity forming transitions to *F. truncicola.* According to Forel, the hairs are like those of *truncicola* but the color, though variable according to the formicaries, tends to become more like that of *pratensis;* the hairs may also become shorter and less black.

The females of this form are said to be very rare (Wasmann). The nests, according to Forel, have a structure intermediate between those of *pratensis* and *truncicola.*

27. F. RUFA AGGERANS Wheeler.

F. rufa McCook, Proc. Acad. nat. sci. Phil., 1884, p. 57–65.

F. rufa subsp. *obscuriventris* Mayr, var. *rubiginosa* Emery, Zool. jahrb. Syst., 1893, **7**, p. 644, 650, ☿ (nec ♀).

F. rufa subsp. *obscuripes* var. *rubiginosa* Wheeler, Ants, 1910, p. 202, 570, fig. 111.

F. rufa aggerans nom. nov., Wheeler, Psyche, 1912, **19**, p. 90.

WORKER. Length 3.5–8.5 mm.

Head and thorax opaque, finely shagreened; mandibles delicately striated, smoother and more shining towards their bases, clypeus finely longitudinally striated. Frontal area subopaque, less shining than in the European forms. Gaster and legs opaque.

Clothed with suberect, grayish or yellowish hairs, abundant on the head, gula, thorax, gaster, and fore coxae, sparser on the tibiae. Eyes hairy. Pubescence on the gaster very fine and dense so that the character of the surface is concealed and this region has a grayish caste.

Head, thorax, and petiole rather bright red, in large specimens immaculate or with only a faint clouding of brown on the divisions of the pro- and mesonotum in smaller workers; legs and antennae dark red or brown, scapes usually paler; in small workers the whole surface of the body may be brown, with the mandibles, clypeus, and anterior portion of the head more reddish; medium sized workers intermediate in having the pro-, meso-, and epinotum, and the petiole more or less infuscated.

FEMALE. Length 8–9 mm.

Surface of body slightly smoother and more shining than in the worker. Gaster, especially, more shining.

Pilosity as in the worker but the hairs longer and somewhat sparser. Pubescence on the gaster in fresh specimens rather dense and concealing the very shining surface but apparently very easily rubbed off. In such specimens the gaster is as smooth and shining as in the European *rufa.*

Head and thorax yellowish red, gaster black; posterior portion of head, front, middle of clypeus, borders of mandibles, posterior border of pronotum, remainder of thorax, except in some specimens, the anterolateral corners of the mesonotum, upper portion of petiole, legs, and antennae, dark brown. Wings slightly infuscated, with brown veins and stigma.

MALE. Length 7–9.5 mm.

Body opaque. Head and thorax more coarsely shagreened than in the European *rufa*. Frontal area somewhat shining.

Hairs and pubescence gray, not black as in the typical *rufa* and more abundant on all parts of the body. Pubescence longer and denser on the legs and gaster so that this region is not shining. Tibiae with sparse, oblique hairs. Eyes hairy.

Deep black throughout, including the legs; genital sclerites yellowish at their bases. Wings as in the female.

HOST (TEMPORARY). Unknown, probably one of the boreal forms of *F. fusca*.

TYPE LOCALITY.— Colorado.

Colorado: Florissant, Lake George, Boulder, Malvern, Salida, Denver, Colorado Springs, Buena Vista (Wheeler); Breckenridge, West Cliff (P. J. Schmitt); Fort Collins (C. P. Gillette); Ute Pass, South Park, Leadville (H. C. McCook); Steamboat Springs (T. D. A. Cockerell).

Montana: Nigger Hill, Powell County (W. M. Mann).

New Mexico: Pecos (T. D. A. Cockerell); Beulah (Mrs. W. P. Cockerell); Barela Mesa (Miss Anna Gohrman).

Utah: Lehi (W. A. Hooker).

Texas: Fort Davis (Wheeler).

Idaho: Lewiston, Market Lake, Collins, and Moscow (J. M. Aldrich).

Wyoming: Carbon County.

North Dakota: Jamestown (H. C. McCook).

Alberta; Medicine Hat (J. C. Bradley).

Emery cites this form from Nebraska, Colorado, and Dakota but I prefer to regard Colorado as the type locality. The female mentioned by Emery from Louisiana probably does not belong here. Emery's description is unfortunately very brief and he does not sufficiently differentiate this form from *obscuripes*. Forel states emphatically that *obscuripes* has only pubescence on its tibiae. I find that all specimens of *aggerans* have prominent oblique hairs on the tibiae, though sometimes few in number. This and the opaque, more pubescent, grayish gaster, and much more abundant pilosity on the body in general serve to distinguish the worker of *aggerans*.

F. aggerans is undoubtedly the common "thatching ant" of the Western States. It constructs a nest very much like that of *F. rufa pratensis* in Europe, at altitudes varying from 6,000–8,000 ft. The vegetable débris used in the construction of the mounds is often very coarse. McCook mentions this ant or possibly the true *obscuripes* as occurring at Iowa Gulch near Leadville, Colo., at an elevation of 11,300 ft.

28. F. RUFA AGGERANS var. MELANOTICA Emery.

F. rufa obscuriventris var. *melanotica* Emery, Zool. jahrb. Syst., 1893, **7**, p. 644, 650, ☿; Wheeler, Ants, 1910, p. 570.

WORKER. Length 4–8 mm.

Differing from the worker of the typical *aggerans* in color and pubescence. The thorax of even the large workers has a strong tendency to infuscation, so that in some colonies such individuals are black with a red head, which is sometimes clouded with brown in the ocellar, occipital, and frontal region. The pilosity is the same as in the typical *aggerans*, but the pubescence on the gaster is more dilute, so that this region is more shining and the shagreened surface is visible.

FEMALE. Length 8 mm.

Resembling the female of *aggerans*, but the infuscation of the thorax is more extensive, involving also the pronotum. Gaster very smooth and shining.

MALE. Length 8 mm.

Differing from the male of *aggerans* only in having the gaster more shining, owing to the sparse pubescence. Frontal area scarcely shining. Eyes hairy.

TYPE LOCALITY.— Wisconsin.

Wisconsin: Dodges' Corner, Waukesha Co., Waupaca (C. E. Brown); Prairie du Chien (H. Muckermann).

Illinois: Rockford (Wheeler); Algonquin (W. A. Nason).

South Dakota: Harding County (S. S. Visher).

Nebraska (Willy).

Wyoming: Medicine Bow (F. M. Chapman).

Oregon: (Amer. Mus. Nat. Hist. Coll.).

Washington: Olympia (T. Kincaid); Pullman (W. M. Mann); Puget Sound (Leconte).

British Columbia: Vernon (W. H. Britton).

I have described the female and male from specimens taken at Pullman, Wash., by Mr. W. M. Mann. I have seen the nests of this form only in Illinois. These are much smaller than the nests of the typical *aggerans,* being rarely more than a foot or 18 inches in diameter. They are built of coarse materials in open grassy fields. Apparently *melanotica* in its deeper pigmentation and its fondness for such situations and for lower altitudes bears about the same relation to *aggerans* that *F. pratensis* does to the typical *rufa* in Europe.

29. F. RUFA OBSCURIPES Forel.

F. rufa st. *obscuripes* Forel, Ann. Soc. ent. Belg., 1886, **30**, C. R. p. xxix; Ibid., 1904, **48**, p. 152, ☿; Wheeler, Ants, 1910, p. 570.

F. rufa obscuriventris Mayr var. *obscuripes* Emery, Zool. jahrb. Syst., 1893, **7**, p. 644, 650, ☿.

WORKER. Length 3.8–8 mm.

Similar to the typical *rufa* of Europe, but the large individuals have the head and thorax of a lighter red and entirely or almost entirely without dark spots on the head and thorax, whereas the legs and petiole are blackish brown or reddish brown. The small workers are of a much darker color and have the head and thorax spotted with brown. Gaster subopaque, deep brown or blackish, covered with slightly longer and denser, gray pubescence than the typical *rufa,* while the erect hairs on the gaster, head, and thorax are rather sparse, inconspicuous and less numerous than in the true *rufa* and the subspecies *pratensis.* Tibiae without erect or suberect hairs and covered merely with appressed pubescence. Gula with a few erect hairs. Eyes hairy.

HOST (TEMPORARY). Unknown; probably one of the boreal forms of *F. fusca.*

TYPE LOCALITY.— Wyoming: Green River (S. H. Scudder).

Wyoming: Elk Creek (R. P. Currie).

Montana: (Amer. Mus. Nat. Hist. Coll.).

Washington: Loon Lake (S. Henshaw); Rock Lake (A. L. Melander).

Colorado: Boulder (Wheeler).

Arizona: Thatcher (R. V. Chamberlin).

British Columbia: Golden (W. Wenman); Summerland (W. H. Britton).

This form is imperfectly known. Forel insists on regarding it as a subspecies, and he may be right, but it should be pointed out that the absence of erect hairs on the tibiae is perhaps not as strong a character as he supposes. One often finds workers of what I regard as Emery's *rubiginosa* (*aggerans*) that have very few suberect hairs on the tibiae.

The specimens from Boulder, Colorado, represent a series from a single colony, the largest with a few suberect hairs on the tibiae, the medium sized and small workers without any, so that one is in doubt as to which subspecies they belong. I have not seen females and males of the true *obscuripes* and am inclined to believe that further study may show that both *obscuripes* and *aggerans* are really the same rather variable subspecies. Both build the same type of nest, a dome-shaped mound of twigs and other vegetable débris, often very coarse and very much like the nests of the European *pratensis* in size and shape. *Formica obscuripes,* like the typical *aggerans,* is peculiar to British Columbia and the Northwestern States, being best represented at altitudes between 5,000 to 8,000 ft.

30. F. RUFA OBSCURIPES var. WHYMPERI Forel.

F. rufa st. *obscuripes* var. *whymperi* Forel, Ann. Soc. ent. Belg., 1904, **48**, p. 152, ☿.
F. rufa obscuripes var. *whymperi,* Wheeler, Ants, 1910, p. 570.

WORKER. With the color and aspect of the darker forms of *F. pratensis* of Europe; front, vertex, occiput, and dorsum of pronotum and mesonotum blackish; with the same pubescence and sculpture, but with the sparse pilosity of *obscuripes;* tibiae without suberect hairs.

TYPE LOCALITY.— British Columbia: Vermillion Pass, 5,000–6,500 ft. (E. Whymper).

This form seems to have been described from a single worker. I have not been able to recognize it among the material collected in British Columbia by J. C. Bradley and W. Wenman.

31. F. TRUNCICOLA TRUNCICOLA Nylander.

F. truncicola Nylander, Acta Soc. Fennica, 1846, **2**, p. 907, ☿ ♀; Ibid., 1849, **3**, p. 26, 29, ♂; Förster, Hymen. stud., 1850, **1**, p. 21, ♀ ♂ (nec ☿); Schenck, Jahrb. Ver. nat. Nassau, 1852, **8**, p. 33, 139, 145, ☿ ♀ ♂; Stettin. ent. zeit., 1853, **14**, p. 160; Mayr, Verh. Zool. bot. ver. Wien, 1855, **5**, p. 334, ☿ ♀ ♂; Europ. Formicid., 1861, p. 46, 48, ☿ ♀ ♂; Forel, Bull. Soc. Vaud. sci. nat., 1875, ser. 2, **14**, p. 58; Mayr, Fedtschenko's Turkestan. Formicid., 1877, p. 6; Ern. André, Spec. Hymen. Europe, 1882, **2**, pt. 14, p. 183, 187, 189, ☿ ♀ ♂; Dalla Torre, Catalog. Hymen., 1893, **7**, p. 213; Bingham, Fauna Brit. Ind., 1903, **2**, p. 334; Forel, Ann. Mus. St. Petersbourg, 1904, **8**, p. 385; Ruzsky, Formicar. Imper. Ross., 1905, p. 330, fig. 63, 64.

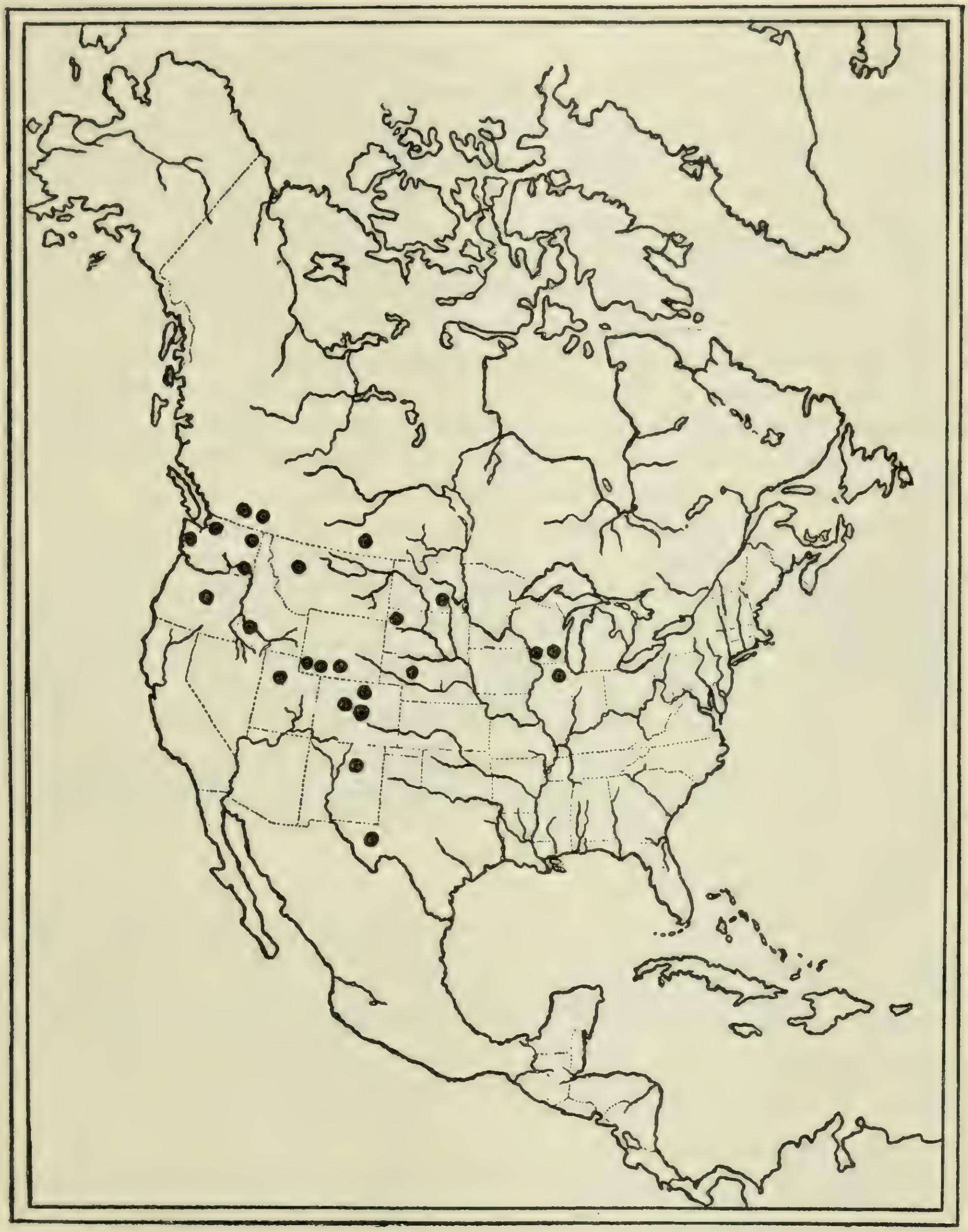

Fig. 2.— Distribution of the Nearctic forms of *Formica rufa*.

F. rufa st. *truncicola* Forel, Denks. Schweiz. gesell. naturw., 1874, **26**, p. 52, ☿ ♀ ♂.
F. rufa subsp. *truncicola* Emery, Deutsch. ent. zeitschr., 1909, p. 187.

WORKER. Length 3.5–8.5 mm.

Closely resembling *F. rufa* in form, but pro- and mesonotum often somewhat less convexed and rounded. Petiole broad, compressed anteroposteriorly, with sharp border, often distinctly notched above. Body, including the gaster, opaque, finely shagreened; mandibles, clypeus, frontal area, and in large workers the anterior part of the head, shining; mandibles finely and superficially striated.

Hairs short, golden yellow, very abundant, covering the body and legs; antennal scapes also often with some oblique or suberect, short hairs. Eyes hairy. Pubescence very short and rather sparse.

Bright red or yellowish red; funiculi and tibiae brown; gaster, with the exception of the anal region and a large yellowish red spot at the base, brownish black. In small workers the vertex and a spot on the pro- and mesonotum are brown, and occasionally the vertex may have a brown spot in large individuals.

FEMALE. Length 8–9 mm.

In sculpture, pilosity, and color very similar to the worker. A spot on the vertex, two or three longitudinal stripes on the mesonotum, the scutellum and gaster, with the exception of the basal half of the first segment, tibiae, and antennal funiculi brown. More rarely the head and thorax, with the exception of the scutellum, are entirely red; sometimes each of the gastric segments is red at the base, at least on the ventral side. Gaster less shining than in the typical *rufa*. more shining than in *pratensis*. Hairs abundant, delicate, yellow, varying considerably in length. Wings infuscated towards their bases, with brown veins and stigma.

MALE. Length 7–9 mm.

Differing from the male of *rufa* and *pratensis* in being much more hairy. Hairs and pubescence yellowish or grayish, the latter rather long on the gaster, legs, and funiculi. Eyes hairy. Frontal area shining. Head, thorax, and antennae black; petiole and gaster often more brownish black; tips of mandibles, genitalia, and legs, except their coxae, reddish yellow. Wings colored as in the female.

HOST (TEMPORARY). *F. fusca*.

North and Middle Europe, Alps, Caucasus, Siberia, Turkestan, Cashmir; Lahoul, on the frontier of Thibet, Eastern Buchara; Island of Sachalin.

Although Forel, Emery, and several other recent authors have regarded *F. truncicola* as a mere subspecies of *rufa*, it seems to me to rank as an independent species. This has, indeed, been the view of most of the older authorities, as will be seen by consulting the synonymy, and Emery himself says: "*F. rufa truncicola* dürfte eher als besondere Art betrachtet werden." The difference is more apparent in the habits, perhaps, than in structure, for *truncicola* does not build large independent mound-nests like *F. rufa, pratensis, aggerans, obscuripes* and their varieties, but nests about stumps and logs or the roots of plants, though it banks these with vegetable detritus. The same habit holds of the forms which I regard as American subspecies and varieties of *truncicola.*

Like *rufa*, the *F. truncicola* queen establishes her colony by temporary parasitism on *F. fusca,* as Wasmann has shown.

32. F. TRUNCICOLA TRUNCICOLA var. YESSENSIS Forel.

F. rufa race *truncicola* var. *yessensis* Forel, Mitth. Naturh. mus. Hamburg, 1901, **18**, p. 66, ☿.

F. rufa truncicola var. *yessensis* Ruzsky, Formicar. Imper. Ross. 1905, p. 335; Emery, Deutsch. ent. zeitschr., 1909, p. 188.

WORKER. Differing from the typical *truncicola* in having the basal surface of the epinotum somewhat shorter and more convex and in the sparser, shorter pilosity. There are very few hairs on the antennal scapes and none on the extensor surfaces of the tibiae. The flexor surfaces bear the usual series of oblique bristles.

Japan: Serachi, Province Ishikari, Island of Yesso.
Siberia: Tomsk and Tobolsk, according to Ruzsky.

33. F. TRUNCICOLA TRUNCICOLA var. SINENSIS, var. nov.

WORKER. Length 4–8 mm.

Opaque; mandibles and clypeus slightly shining, delicately longitudinally striate; frontal area and frontal groove very smooth and shining.

Erect hairs golden yellow, less abundant than in the typical *truncicola,* absent on the upper surface of the head and clypeus, middorsal region of the pro- and mesonotum and flexor surfaces of the tibiae, long on the gula, epinotum, fore coxae, and gaster. Eyes hairless. Pubescence fine and rather sparse, most easily visible on the epinotum and on the gaster where it is sufficiently abundant to produce a grayish tinge, extremely fine on the antennae, head, pro- and mesonotum.

Head, thorax, petiole, and legs deep, dull red; mandibles and front of head a little darker; cheeks and clypeus a little paler; smallest workers with the upper surface of the head and a spot on the pro- and mesonotum fuscous. Gaster black, anal region, and often also in large individuals, the base of the first segment, dull reddish.

Described from numerous workers from Chung-King, Western China, (Amer. Mus. Nat. Hist. Coll.).

This variety is evidently very close to *yessensis* Forel, but the antennal scapes are very smooth and without any traces of oblique hairs, the color is very deep and the distribution of the hairs is peculiar, especially their absence on the whole upper surface of the head and clypeus and their nearly complete absence on the convex portions of the pro- and mesonotum.

34. F. TRUNCICOLA DUSMETI Emery.

F. rufa dusmeti Emery, Deutsch. ent. zeitschr., 1909, p. 188, ☿ ; Forel, Rev. Suisse Zool., 1911, **19**, p. 457, 458.

Worker. Resembling the typical *truncicola* in its light red coloration; head and thorax red, without spots or with a blackish spot on the front; gaster quite opaque, black, with red basal spot; antennae and legs brown, scapes and femora red. Head and thorax entirely without erect hairs; eyes hairless; gaster covered with rather abundant short hairs.

This subspecies, based on three specimens collected by Dusmet at Peñalosa, in Spain, has recently been taken in Norway by Forel. According to Emery, it is very similar to the North American *F. truncicola integra* Nyl.

35. F. TRUNCICOLA INTEGROIDES Emery.

F. rufa subsp. *obscuriventris* Mayr. var. *integroides* Emery, Zool. jahrb. Syst., 1893, **7**, p. 649, ♀, Wheeler, Ants, 1910, p. 570.

Worker. Length 3.5–8 mm.

Body, including the mandibles and clypeus, opaque. Mandibles densely and sharply striate, with scattered punctures. Frontal area smooth, only slightly shining. Clypeus sharply carinate. Thorax and petiole much as in the typical *truncicola*, but base of epinotum somewhat more convex.

Pubescence yellowish, dense, distinct on the head and thorax, long and conspicuous on the gaster, sparse on the legs. Hairs golden yellow, very sparse on the upper surface of the head, thorax, and petiole, more abundant on the gula, short and appressed on the extensor surfaces of the tibiae. Eyes hairy.

Light yellowish red; mandibles, legs, and antennae darker, femora more brownish; gaster dark brown, with yellowish anal region. Small workers sometimes have the top of the head and thorax spotted with pale brown.

FEMALE. Length 8 mm.

Head, excluding the mandibles, as broad as long, with very straight posterior and lateral borders, the latter strongly converging anteriorly; posterior corners of head rather sharp. Antennal scapes reaching about twice their greatest diameter beyond the posterior corners.

Sculpture, pilosity, and color as in the worker. Petiole, thoracic dorsum, and base of gaster with a number of pale, erect hairs; pubescence on the head and thorax even longer and more conspicuous than in the worker. Gaster not shining. Mesonotum with three elongate fuscous spots; funiculus, except its base, the metanotum, and posterior portion of scutellum, blackish. Wings opaque gray, with brown veins and stigma.

HOST (TEMPORARY). Unknown.

TYPE LOCALITY.— California: Coastal mountains.

California: San Gabriel Mountains near Claremont (C. F. Baker); Felton, Santa Cruz Mountains 300–500 ft.; Santa Cruz Beach, Giant Forest (J. C. Bradley); Loma Prieta, Santa Cruz Mountains 3,800 ft. (V. L. Kellogg); King's River Canyon (H. Heath); Corte Madera Creek (W. M. Mann); Pine Lake (J. D. Johnson).

I have redescribed the worker of this form from cotype specimens given me by Mr. Pergande. Although, as Emery states, it is allied to *integra,* it is not a variety of *obscuriventris,* as he believed. He records it from California and Nebraska, but I have seen the form only from the coastal mountains of the former state and regard this as the type locality. It is replaced to the eastward in the Rocky Mountains by the two closely allied varieties described below, which differ from it mainly in pilosity.

According to a statement (*in litteris*) of Prof. Harold Heath, *F. integroides* inhabits open woods and accumulates large quantities of vegetable detritus about the stumps and logs in which it nests. In these particulars its habits are very similar to those of the European *truncicola* and our eastern subsp. *integra.*

F. TRUNCICOLA INTEGROIDES var. COLORADENSIS, var. nov.

F. rufa subsp. *integra* var. *coloradensis* Wheeler, Ants, 1910, p. 570.

WORKER. Length 4–9 mm.

Differing from the typical *integroides* in its somewhat greater average size, in having more shining mandibles and frontal area, and in the pilosity, which is pale yellowish and as abundant as in the European *truncicola,* covering all parts of the body, except the antennae. The scapes often have a few scattered suberect hairs and the eyes are distinctly hairy. Oblique hairs on the extensor surfaces of the tibiae as long as those on the flexor surfaces. The pubescence is also long and abundant, conspicuous on the head and thorax as well as the gaster. Small and large workers are of the same color.

Head, thorax, petiole, legs, and antennae bright red, mandibles darker; gaster dark brown, with red anal region and often with a small red spot at the base of the first segment.

FEMALE. Length 8–10 mm.

Mandibles more opaque and more coarsely sculptured than in the worker.

Pilosity and pubescence similar to those of the worker, but the former whitish, more delicate and less conspicuous on the thorax. Pubescence on the gaster more dilute so that this region is slightly lustrous or shining and not opaque as in the worker.

Color like that of the worker; mesonotum with three elongate brown spots; funiculi, metanotum, and posterior border of scutellum infuscated; mandibles deep red. Wings grayish hyaline, distinctly infuscated towards the base.

TYPE LOCALITY.— Colorado: Florissant, 8,100 ft.

Colorado: Wild Horse and Woodland Park, 8,500 ft. (Wheeler); Ward, 9,000 ft. (T. D. A. Cockerell); Boulder, Breckenridge (P. J. Schmitt).

New Mexico: Pecos, Beulah, 8,000 ft. (T. D. A. Cockerell and Mrs. W. P. Cockerell).

Idaho: Blackfoot, Market Lake (J. M. Aldrich).

Of all our forms this is most like the typical European *truncicola* in pilosity. It differs, however, in color, the red parts being lighter and the gaster with an inconspicuous yellow base and a peculiar bluish bloom, due to the dense gray pubescence covering a blackish surface. Its habits are similar to those of the European species since it nests under and in stumps and logs, filling their interstices with vegetable débris, but the colonies are much larger than those of the European type and, according to my observations, are confined to pine woods and to higher altitudes. The queens were taken in the nests during the latter half of July.

36. F. TRUNCICOLA INTEGROIDES var. HAEMORRHOIDALIS Emery.

F. rufa subsp. *integra* var. *haemorrhoidalis* Emery, Zool. jahrb. Syst., 1893, **7**, p. 652, ☿ ; Wheeler, Ants, 1910, p. 570.

WORKER. Length 4–9 mm.

In structure, sculpture, and color like the preceding variety and the typical *integroides*, but differing from both in having no erect hairs on the head, thorax, petiole, and legs, with the exception of a few on the clypeus and the row of bristles on the flexor surfaces of the tibiae. Suberect hairs on the gaster sparse. The pubescence is the same as in the two preceding forms and the gaster has the same appearance of being covered with a bluish bloom. Eyes hairless. Mandibles and frontal area rather shining. The small workers do not differ in color from the large ones.

FEMALE. Length 9–10 mm.

Similar to the female of *coloradensis* but lacking the erect hairs on the head, thorax, petiole, and legs and the dark spots on the thorax. Wings grayish hyaline, infuscated towards the base, with brown veins and stigma.

MALE. Length 8 mm.

Mandibles rather broad, edentate. Petiole thick, but with a compressed, thin margin, broadly excised in the middle.

Head and thorax opaque; gaster glossy; frontal area slightly shining. Body and legs covered with grayish pubescence which is longest on the thorax and gaster. The erect hairs are very short, but moderately abundant on the head, thorax, and gaster. Tibiae with short, scattered, suberect hairs.

Head, thorax, gaster, and antennae black; tips of mandibles, genitalia and legs testaceous or yellowish, femora and tarsi more or less infuscated. Wings as in the female.

TYPE LOCALITY. — Colorado.

Colorado: Florissant 8,100 ft., Woodland Park 8,500 ft., Ute Pass, Manitou, Garden of the Gods (Wheeler).

Dakota (Emery).

Idaho: Moscow Mts. (J. M. Aldrich).

Nevada: Ormsby County (C. F. Baker).

Washington: Yakima River opposite Ellensburg (S. Henshaw).

This variety lives in the same localities and builds nests very similar to those of the var. *coloradensis.* It is in fact, merely a hairless variety of *integroides* and not a variety of *integra,* which is confined to the Eastern States and has a different kind of pubescence and a very hairless male. The typical *integroides,* and the vars. *coloradensis* and *haemorrhoidalis* have exactly the same macroscopical appearance and differ essentially only in pilosity. They represent in the west the two eastern subspecies *integra* and *obscuriventris.*

37. F. TRUNCICOLA MUCESCENS, subsp. nov.

WORKER. Length 3.5–7 mm.

Head large, excluding the mandibles, about as broad as long, a little broader behind than in front, with straight or very feebly excised posterior border and slightly convex sides. Mandibles 8-toothed. Clypeus strongly carinate and convex at base, its anterior border broadly rounded, not angular. Frontal carinae moderately diverging. Antennae slender, basal funicular joints longer and more slender than the penultimate joints. Pro- and mesonotum not very convex, the mesoëpinotal impression rather shallow, the epinotum with subequal base and declivity, the former convex, the latter feebly concave. Petiole rather thick and narrow, cuneate in profile, convex in front and behind, with rather sharp superior border, which is entire and somewhat angularly rounded in the middle.

Subopaque; mandibles, clypeus, front, and sides of head more shining, the mandibles finely striatopunctate. Frontal area very smooth and shining. Gaster opaque.

Hairs bright golden yellow, short and very sparse on the front, more abundant on the pro- meso- and epinotum, gula, petiolar border, and gaster; absent on the scapes and legs. Pubescence grayish, very dense and rather long on the gaster, well developed but sparser on the head and thorax, especially on the epinotum; very sparse and inconspicuous on the legs. Eyes not hairy.

Light red; mandibles, petiole, antennae, and legs darker and more brownish red; gaster dark or blackish brown, but appearing drab on account of the pubescence; anal region red. Small workers with the upper surface of the thorax and sometimes also the vertex, more brownish red.

FEMALE. Length 6.5–8 mm.

Body small. Head broader than the thorax; eyes rather small. Petiole much as in the worker but with blunter border.

Mandibles, frontal area, and legs shining, remainder of body more opaque than in the worker.

Erect hairs lacking on the body, except on lower surface of petiole, venter, and tip of gaster. There are occasionally a few hairs on the front. Tibiae with oblique and rather long pubescence on their flexor surfaces. Pubescence gray, long, dense, and suberect on the gula, upper surface of the head and thorax, appressed on the antennal scapes and gaster. The latter region is slightly more lustrous than in the worker.

Head, thorax, and petiole sordid brownish yellow; mandibles, ocellar region, posterior corners of head, posterior border of pronotum, mesonotum, scutellum, metanotum, mesopleurae, legs, antennae, and sometimes also the upper border of the petiole, brown; gaster blackish brown, with slightly reddish anal region. Wings distinctly infuscated, scarcely paler towards their tips; veins and stigma brown.

MALE. Length 7–8 mm.

Mandibles edentate, clypeus convex, carinate, transversely impressed behind; head rather broad; eyes large. Petiole low and thick with rounded, entire superior border.

Mandibles and frontal area shining; head and thorax opaque; gaster, pleurae, legs, and genitalia lustrous.

Hairs yellowish, very short, erect, and rather abundant on the head and thorax, sparser and more appressed on the upper surface of the gaster, absent on the legs. Pubescence grayish, moderately developed, most distinct on the gaster.

Black; even the tips of the mandibles not paler; genital sclerites black or castaneous, with yellowish insertions. Wings infuscated as in the female.

TYPE LOCALITY.— Colorado: Colorado Springs. (Wheeler).

Colorado: Colorado City, Malvern, Wild Horse, Manitou (Wheeler); West Cliff (P. J. Schmitt).

This form is rather puzzling. It is perhaps a distinct species, but I have preferred to regard it provisionally as a subspecies of *truncicola.* The small, almost hairless and peculiarly colored females enable one to recognize the species better than the workers, which at first sight resemble those of *F. truncicola integroides* vars. *haemorrhoidalis* and *coloradensis.* The new form differs from these varieties, however, in pilosity and pubescence, and in the smaller average size of all three phases. The males are peculiar in being almost entirely black, even to the greater portion of the genital appendages.

I found several colonies of this ant both in 1903 and 1906, each containing many females and males. They were nesting in open places, at altitudes of about 5,000–7,000 feet, under stones banked with vegetable detritus.

38. F. TRUNCICOLA INTEGRA Nylander.

F. integra Nylander, Ann. sci. nat. Zool., 1856, ser. 4, **5**, p. 62 *nota*, ☿; F. Smith, List Brit. anim. Brit. mus., 1858, pt. 6, p. 54, ☿; Dalla Torre, Catalog. Hymen., 1893, **7**, p. 200.

F. integra var. *similis* Mayr, Verh. Zool. bot. ver. Wien, 1886, **36**, p. 425, ☿ ♀ ♂.

F. rufa subsp. *integra* Emery, Zool. jahrb. Syst., 1893, **7**, p. 652, pl. 22, figs. 4, 8; Wheeler, Bull. Amer. mus. nat. hist., 1906, **22**, p. 67; Ants, 1910, p. 204, 570, fig. 112.

WORKER. Length 4–8 mm.

Closely resembling the worker of *F. truncicola integroides* var. *haemorrhoidalis* in lacking the pilosity on the head, thorax, petiole, and legs, but the petiole is broader and with a sharper border, the surface of the body is opaque, except the mandibles, frontal area, and gaster which are all somewhat shining. Mandibles sharply, clypeus less distinctly striated.

Pubescence very fine, visible on the head, thorax, and appendages, but sparse on the gaster so that the surface is visible. Hairs on the gaster very sparse, often more abundant on the venter and tip. Eyes hairless.

Head, thorax, petiole, and appendages bright red, mandibles darker. Gaster black, only the anal region and extreme base of first segment slightly tinged with red. The red portions of small workers rarely somewhat darker above than in large individuals.

FEMALE. Length 8–9 mm.

Closely resembling the worker in sculpture, color, and pilosity. Pubescence on gaster denser so that this region is subopaque or but slightly shining. Mesonotum immaculate or with very faint brownish indications of the three spots. Metanotum black, scutellum, with the exception of its anterior border, infuscated. Wings grayish hyaline, infuscated at the base, with brown veins and stigma.

MALE. Length 7–8 mm.

Mandibles broad, sometimes indistinctly dentate. Eyes rather large. Petiole compressed, with rather sharp, broadly excised superior border.

Head and thorax opaque, frontal area strongly, gaster and mandibles more feebly, shining.

Erect hairs absent on head, thorax, gaster, and appendages. Pubescence grayish, rather abundant but fine, most conspicuous on the thorax, legs, and gaster. Eyes hairless.

Black; antennae dark brown; tips of mandibles reddish. Legs and genitalia yellow, coxae infuscated. Wings somewhat more heavily and uniformly infuscated than in the female.

Host (Temporary). *F. fusca* var. *subsericea.*

Maine: Lower Goose Island, Casco Bay (Wheeler); Monmouth (Frost).

New Hampshire: Littleton, Hannover (C. M. Weed); Durham (W. F. Fiske); Mt. Moosilauke, 1,700 ft. (Wheeler).

Massachusetts: Wellesley (A. P. Morse), Forest Hills (Wheeler); W. Springfield (Geo. Dimmock).

Connecticut: Colebrook (Wheeler).

New York: High Bridge; West Farms (J. Angus); Ashokan, Bergen Beach (G. v. Krochow); Mosholu, Bronxville (Wheeler); Long Beach, Long Island.

New Jersey: Jamesburg, Lakehurst (Wheeler); Clementon.

Pennsylvania: Enola, Frankford, Lawndale; West Chester (J. C. Bradley).

North Carolina: Black Mts. (W. Beutenmüller).

Georgia: Atlanta, Stone Mt. (J. C. Bradley).

Indiana: Camelton, and Wyandotte (W. S. Blatchley).

Illinois: Rockford (Wheeler).

South Dakota; Hill City (Th. Pergande).

Michigan: Isle Royale (H. A. Gleason).

Alabama: Cullman (P. J. Schmitt).

Nova Scotia: Bedford (W. Reiff); Digby (J. Russell).

The specimens from northern localities like Nova Scotia, Isle Royale, and Northern Illinois have the red parts darker and somewhat clouded with fuscous and the pubescence on the gaster even sparser than in the typical form so that this region is more shining, but with the material on hand it hardly seems advisable to regard these as representing a distinct variety.

This ant, which occupies similar stations in the Eastern States to those inhabited by *integroides* in the West, has also very similar habits, living in huge colonies in rather rich, open woods in hilly regions. The nests are built in stumps and logs and under stones, and resemble those of the European *truncicola,* but are on a larger scale. A single colony often occupies a number of nests covering a considerable area. The winged forms make their appearance in August.

39. F. TRUNCICOLA OBSCURIVENTRIS Mayr.

F. truncicola var. *obscuriventris* Mayr, Verh. Zool. bot. ver. Wien, 1870, **20**, p. 951, ☿; Ibid., 1886, **36**, p. 426; Dalla Torre, Catalog. Hymen., 1893, **7**, p. 214.

F. rufa subsp. *obscuriventris* Emery, Zool. jahrb. Syst., 1893, **7**, p. 649; Wheeler, Ants, 1910, p. 570.

F. dryas Wheeler, Bull. Amer. mus. nat. hist., 1905, **21**, p. 268, ☿ ♀.

WORKER. Length 3.5–7.5 mm.

Averaging smaller than the preceding subspecies. Base of epinotum usually convex and rounded. Petiole moderately broad, strongly compressed anteroposteriorly, its dorsal border thin and sharp, entire and produced upward in the middle.

Opaque; mandibles and clypeus slightly shining, longitudinally striate, frontal area very smooth and shining. Gaster subopaque or slightly shining, finely shagreened and punctate.

Hairs and pubescence yellowish, the former short, erect, as abundant as in the typical *truncicola*, covering all parts of the body except the antennae; scapes sometimes with a few scattered hairs on their posterior surfaces. Eyes hairy. Pubescence distinct and rather dense on the head and thorax, sparser and finer on the gaster so that the surface is clearly visible.

Deep red, when mature; mandibles and corners of clypeus darker; gaster black, with the anal region and sometimes also a small spot at the base of the first segment reddish. Large workers very rarely, smallest workers somewhat more frequently, with a brownish cloud in the region of the ocelli and on the pro- and mesonotum.

FEMALE. Length 7–8 mm.

Surface of body more shining than in the worker, especially the gaster, which is as glabrous and shining as in the female of the typical *rufa*. Mandibles coarsely striatopunctate, head and thorax very finely and densely punctate; clypeus, front, and anterior portion of mesonotum with several large, elongate punctures.

Hairs longer and more flexuous than in the worker, especially on the top of the head, gula, thoracic dorsum, and tibiae, varying greatly in length, sparse on the pleurae and absent on the gaster, except on the ventral surface and tip. Antennal scapes often with a row of long, flexuous, erect hairs along their posterior surfaces. Eyes hairy. Pubescence yellowish, abundant and rather dense on the thorax and legs; sparse on the gaster.

Deep red, three elongate spots on the mesonotum, the posterior portion of the scutellum, the metanotum, and the gaster black. Tibiae, tarsi, and funiculi dark brown. Anal region sometimes reddish; first segment often with a yellowish red spot at the base. Wings strongly infuscated, with paler tips, brown veins and stigma.

MALE. Length 8 mm.

Mandibles edentate, rather narrow. Eyes large. Petiole more compressed anteroposteriorly than in the other subspecies, its border rather sharp and not excised.

Pilosity and pubescence yellowish, abundant, the former suberect, absent on the upper surface of the gaster, which is covered with rather long, appressed pubescence. Legs covered with short, suberect hairs. Eyes hairy.

Head and thorax, including the frontal area, opaque; gaster somewhat shining.

Black; legs and genitalia yellow, coxae and bases of the femora dark brown. Wings infuscated as in the female.

HOST (TEMPORARY). *F. fusca* var. *subsericea.*

TYPE LOCALITY.— Connecticut: (Mayr).

Massachusetts: Stony Brook Reservation, Blue Hills, near Boston, Ellisville (Wheeler); Wellesley (A. P. Morse).

Maine: Sebascodegan Island, Casco Bay (Wheeler).

New York: Saugerties (G. v. Krochow); Ithaca (Cornell Univ. Coll.).

New Jersey (Mayr).

District of Columbia: Washington (A. Forel).

Virginia (Mayr).

Indiana: Culver, Tippecanoe Lake (W. S. Blatchley).

Illinois: Rockford (Wheeler).

Wisconsin: White Fish Bay, near Milwaukee (Wheeler).

Colorado: Florissant (Wheeler); Flagstaff Mt., Boulder (T. D. A. Cockerell).

British Columbia: Carbonate, Ravelstoke (J. C. Bradley); Golden (W. Wenman).

Ontario: Toronto (R. J. Crew).

This ant was originally described from Connecticut, but in a later paper (1886) Mayr cited it from several of the Atlantic States and also from Colorado, California, New Mexico, and Arizona. As it is very rare in Colorado, and as I have never received it from other Western States, I believe Mayr must have confounded it with specimens of *F. rufa aggerans.* This would be easy for large greasy specimens of *aggerans* are very similar to large workers of *obscuriventris.* In fresh specimens, however, the gaster of the latter has a very different appearance, being much as in *melanotica,* but it is readily distinguished from this form by the uniform deep red color of the head, thorax, petiole, and legs.

F. obscuriventris nests under large stones in open woods, often banking the edges of the stones with vegetable débris. The colonies are much smaller than those of *integra,* and *integroides* and rarely extend over more than one nest. Many queens are retained in the nest and these are often very imperfectly deälated. As early as April 3 I have found many perfectly winged queens as well as several with the wings more or less gnawed off, in various colonies near Boston. These queens had evidently been retained by the maternal colony or adopted after leaving other colonies on their nuptial flight. Tanquary and I have recently shown that the queens of this ant establish new colonies through temporary parasitism on *F. fusca* var. *subsericea.*

40. F. TRUNCICOLA OBSCURIVENTRIS var. GYMNOMMA Wheeler.

F. dryas var. *gymnomma* Wheeler, Bull. Amer. mus. nat. hist., 1905, **21**, p. 269, ☿.

F. rufa subsp. *obscuriventris* var. *gymnomma* Wheeler, Ants, 1910, p. 570.

WORKER. Length 3.5–7.5 mm.

Differing from the typical *obscuriventris* only in having the eyes hairless and in having the erect hairs on the body less abundant, especially on the upper surface of the head.

TYPE LOCALITY.— New York: Cold Spring Harbor, Long Island, (Wheeler).

Massachusetts: Wellesley (Wheeler).
Georgia: Clayton, 2,000–3,700 ft. (W. T. Davis).
Illinois: Rockford (Wheeler).

41. F. URALENSIS Ruzsky.

F. uralensis Ruzsky, Arb. Ges. naturf. Kasan, 1895, **28**, p. 13, ☿ ♀ ♂; Berlin. ent. zeitschr., 1896, **41**, p. 69; Formicar. Imper. Ross., 1905, p. 348, fig. 66; Emery, Deutschr. ent. zeitschr., 1909, p. 189.

WORKER. Length 5–8 mm.

Head as broad as long, but little narrower in front than behind. Frontal carinae strongly diverging. Clypeus strongly carinate, with produced, angular, anterior margin. Antennae robust, the scapes short and thick. Petiole rather broad, compressed anteroposteriorly, with sharp, rounded, entire border.

Opaque; mandibles subopaque, densely striatopunctate. Sides of head glossy. Frontal area opaque.

Pilosity of the whole body very sparse, tibiae without oblique or suberect hairs. Gula with a few erect hairs. Pubescence yellowish, fine and sparse on the gaster and legs, denser on the cheeks and thorax. Eyes hairless.

In color resembling the darkest forms of *F. rufa pratensis*, but the

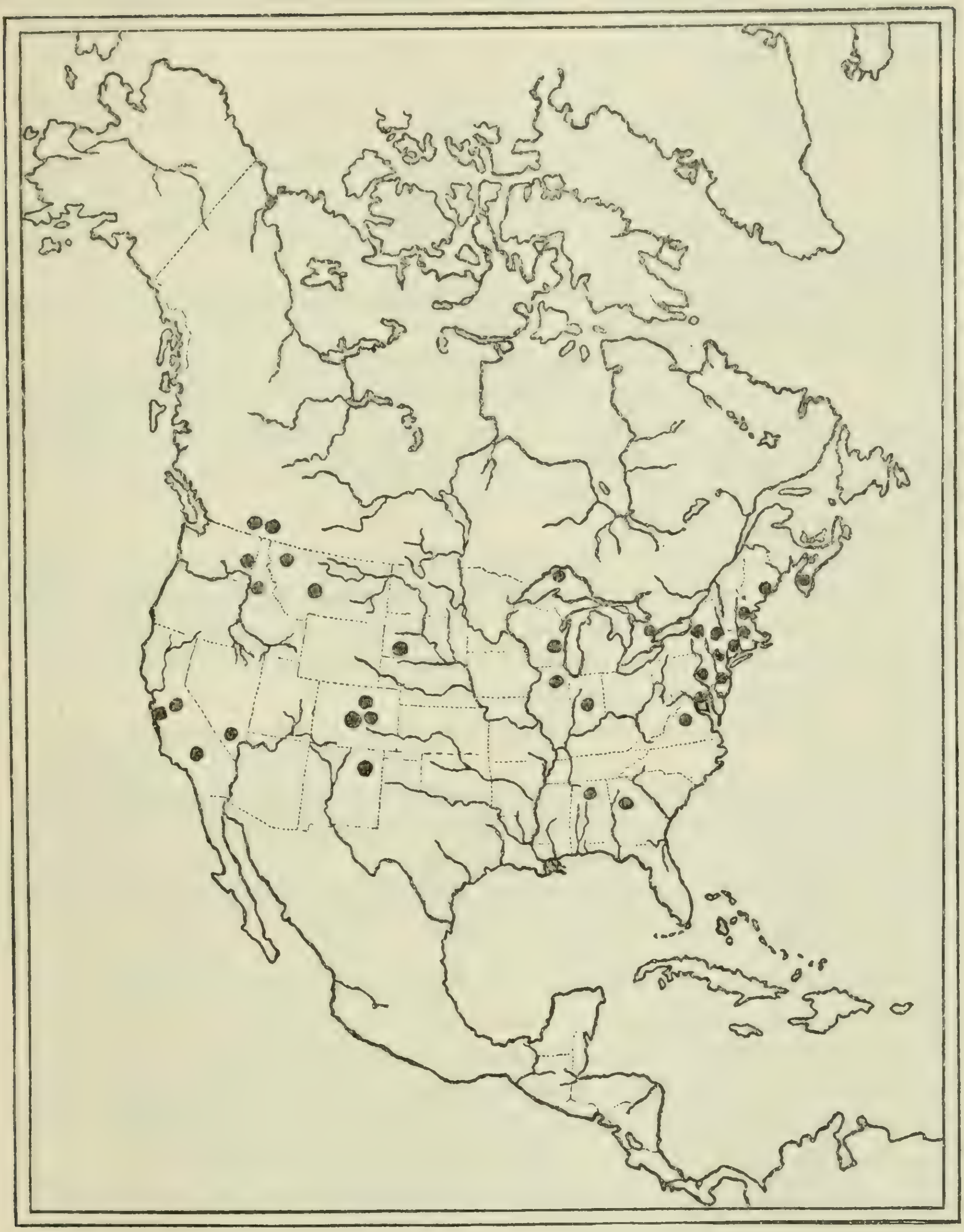

FIG. 3.— Distribution of the Nearctic forms *of Formica truncicola.*

whole of the head black, except the mandibles and a spot on the gula, which are red. Pro- and mesonotum with a black spot as in *pratensis*. Gaster black, with the base of the first segment reddish yellow. Antennae and legs brown.

FEMALE. Length 8.5–10 mm.

Antennae shorter and thicker than in the worker. Head, excluding the mandibles, a little broader than long.

Body densely punctate, head and thorax more coarsely, gaster more finely; head and thorax opaque, except the mandibles, which are coarsely striatopunctate. Clypeus delicately longitudinally striate. Gaster somewhat shining.

Pilosity pale, sparse, and somewhat longer, pubescence even sparser and more indistinct than in the worker.

Dark brown or black, mandibles except their borders, anterior portion of pronotum, inferior pleurae, venter and base of first gastric segment yellowish or reddish. Legs brown or blackish brown, coxae reddish. Wings not infuscated but merely tinged with yellowish at their bases. Veins and stigma brown.

MALE. Length 9–11 mm.

Mandibles tridentate. Head very broad and shorter than in *rufa* and *sanguinea*, eyes rather small, the antennae, and especially their scapes, shorter and much thicker. Petiole high and rather compressed anteroposteriorly, with a rounded superior border, which is scarcely or not at all excised in the middle.

Body, including the mandibles and frontal area, opaque; gaster feebly glossy.

Pilosity and pubescence grayish, the former sparse especially on the head and gaster, most conspicuous on the thoracic dorsum, the pubescence rather long and dense on the gaster.

Body, legs, and antennae black; genital appendages with yellow bases and black tips. Wings colorless.

Siberia, from the middle and southern portions of the Ural Mountains to Transbaikalia.

The nests are described as similar to those of *F. rufa* and *pratensis* and in the Ural Mountains are located on the summits and slopes of hills which are overgrown with grass and scattered birches. The pupae are said by Ruzsky not to be enclosed in cocoons.

The species is easily recognized by the peculiar coloring and robust antennae of all three phases.

42. F. ADELUNGI Forel.

F. adelungi Forel, Ann. Mus. St. Petersbourg., 1904, 8, p. 384, ♂; Ruzsky, Formicar. Imper. Ross., 1905, p. 420, ♂; Emery, Deutsch. ent. zeitschr., 1909, p. 189, ♂.

MALE. Length 7.7 mm.

(After Forel). Differs from all the known species, except *F. sanguinea* in its mandibles armed with 5–6 teeth, which are even more distinct than in the male *sanguinea.* The basal half of the mandibles is, moreover, very narrow, but the terminal half is enlarged. Clypeus carinate, with perfectly entire anterior border, thus distinguishing the species from *sanguinea.* Head very short, more than $1\frac{1}{2}$ times as broad as long, with very large eyes, occupying $\frac{2}{3}$ of its sides. Petiole with its superior border notched in the middle. In other respects like *F. sanguinea.* The same is true of the thorax. Wings shorter than in *F. sanguinea,* not surpassing the gaster and hyaline throughout, with brownish veins and stigma. Color, sculpture, and pilosity in other particulars as in *F. sanguinea* but the legs are brown, the pilosity is sparser (almost absent except on the lower surface of the gaster), and the sculpture and pubescence are a little finer.

Oasis Satsch-zou in the desert of Gobi (Roborovsky and Kozlov).

The affinities of this species, which is known from only a single male specimen, are not at all clear. Forel evidently believed it to be related to *F. sanguinea,* whereas Emery places it near *uralensis.* Its exact position cannot be determined till the worker and female have been discovered.

43. F. FORELIANA, sp. nov.

WORKER. Length 4–6 mm.

Mandibles 8-toothed. Head, excluding the mandibles, a little longer than broad, a little narrower in front than behind, with straight posterior and feebly convex lateral borders. Clypeus strongly carinate throughout its length, with broadly rounded, projecting anterior border. Frontal carinae very slightly diverging behind, nearly parallel. Antennae long and slender, joints 2–4 of the funiculus longer and more slender than joints 8–10. Maxillary palpi very long, decidedly longer than in any of the preceding species of the *rufa* group. Thorax rather long, the pro- and mesonotum not very convex, the mesoëpinotal constriction rather shallow, the epinotum with subequal base and declivity, both straight in profile and meeting at a pronounced angle. Petiole rather narrow, cuneate in profile, with feebly convex anterior and more flattened posterior surface, blunt lateral and rather sharp, rounded, superior margin which is either entire or very feebly notched in the middle. Legs moderately long and slender.

Body including frontal area opaque, very finely shagreened; mandibles somewhat coarsely striatopunctate.

Hairs golden yellow, long, slender, erect, sparse; present only on the mandibles, clypeus, front, vertex, pronotum, gaster, fore coxae, and in a single row on the flexor surface of the femora, tibiae, and tarsi. On the gaster the hairs, very conspicuous in certain lights, are present in three or four rows on each of the segments. Pubescence grayish, very fine, dense on the gaster, somewhat sparser on the head, thorax, scapes, and legs.

Brownish red; front, upper surface of thorax, petiole, and femora, especially the hind pair, darker or infuscated; gaster black.

Described from several specimens taken from two colonies at altitudes of 4,500 and 5,600 ft. in the Huachuca Mountains, Arizona, by Mr. C. R. Biedermann.

At first sight the species closely resembles a small *F. sanguinea rubicunda*, especially in the shape of the thorax and petiole and in pilosity, but it differs in having the anterior border of the clypeus projecting and entire, and in the much longer maxillary palpi, much more slender antennae, and the coloration of the head and thorax.

44. F. CILIATA Mayr.

F. ciliata Mayr, Verh. Zool. bot. ver. Wien, 1886, **36**, p. 428, ♀; Emery, Zool. jahrb. Syst., 1893, **7**, p. 655, pl. 22, fig. 12, ♀; Wheeler, Bull. Amer. mus. nat. hist., 1903, **19**, p. 640, fig. 1, ☿ ♀ ♂.

WORKER. Length 3–8 mm.

Mandibles 8-toothed. Clypeus sharply carinate its entire length, its anterior border broadly rounded, not produced. Head, excluding the mandibles, fully as broad as long, a little narrower in front than behind; occipital border slightly concave, especially in large specimens; posterior corners rounded, sides feebly convex, cheeks long. Frontal carinae distinctly diverging behind. Antennae slender, funicular joints 1–4 longer and more slender than the penultimate joints. Maxillary palpi short, pro- and mesonotum not very convex, mesoëpinotal constriction not very deep, base and declivity of epinotum subequal, forming a distinct obtuse angle with each other, the former in profile straight or feebly convex, the latter slightly concave. Petiole rather narrow, cuneate in profile, with slightly convex anterior and flattened posterior surface; border sharp, rounded on the sides, produced upwards as a blunt point in the middle.

Mandibles finely striatopunctate, feebly shining. Clypeus very finely longitudinally striated, remainder of the body delicately shagreened. Whole body opaque, except the frontal area, which is smooth and shining. The clypeus and even the whole head in the largest workers from some colonies, may be more or less shining.

Hairs golden yellow, short, and erect, those on the clypeus and mandibles rather coarse. Upper surface of head naked, gula with a few erect hairs. Eyes bare. Thorax covered with erect hairs, except the mesonotum, mesopleurae, and basal epinotal surface, which are naked. Petiole below and along its border with a fringe of short hairs, also with a few hairs on its anterior and posterior surfaces. Gaster invested with numerous uniformly distributed, short, suberect, obtuse hairs, which are not longer on the terminal than on the basal segments. Legs without oblique hairs, except the customary row on the flexor surfaces of the tibiae. Pubescence grayish, rather long and sparse on the head, thorax, and legs, on the gaster very dense and concealing the surface.

Head, thorax, and petiole of largest workers rich yellowish red, mandibles and clypeal sutures darker. Gaster brown, but appearing gray on account of the dense pubescence, anal segment and often also the venter and the base of the first segment, yellow. Antennae and legs reddish yellow, funiculus towards the tip, coxae, femora, and often also the tibiae, dark brown. The smallest workers usually have the posterior portion of the head, thoracic dorsum, and border of petiole clouded with black or dark brown. In some small specimens the whole body, excepting the mandibles and anterior portion of the head, is uniformly infuscated.

FEMALE. Length 6–8 mm.

Thorax rather small, somewhat narrower than the head. Petiole broadly rounded, its superior border sharp, but not produced in the middorsal line as in the worker.

Mandibles subopaque, striatopunctate. Body and appendages smooth and shining, especially the head, mesonotum, and scutellum, which are very glabrous.

Pilosity extraordinary, consisting of very long, golden yellow hairs, which have a tendency to curl at their ends. These hairs are absent on the upper surface of the head, the mesonotum, and legs, excepting the coxae. They are long and conspicuous on the mandibles and clypeus, on the latter scattered over the disc and also fringing the anterior border. Gula with long, appressed hairs. Remainder of body, excepting the nude portions above mentioned, covered with long woolly hairs which form a prominent fan-like fringe on the border of the petiole. On the gaster they are long and abundant, appressed overlapping and curled at their tips, so that this region of the body appears opaque, in marked contrast to the head and mesonotum. Antennae and legs covered with minute, inconspicuous pubescence, flexor surfaces of fore femora with flexuous hairs, corresponding surfaces of middle tibiae each with a single row, hind tibiae with two rows of stiff hairs.

Color also unusual; rich reddish yellow throughout; only the terminal half of the funiculus, the mesonotum, the adjacent portion of the scutellum, and the alar insertions black or infuscated. Wings uniformly grayish hyaline; veins and stigma more yellowish gray, the latter not very conspicuous.

MALE. Length 6.5–8 mm.

Mandibles edentate, sharply pointed. Head very short, very broad behind the eyes, very narrow in front, occipital border straight. Clypeus strongly carinate. Maxillary palpi 5-jointed. Thorax robust, broader than the head. Petiole thick, convex anteriorly, more flattened posteriorly, border very blunt, evenly rounded and entire both in profile and when seen from behind.

Mandibles and upper surface of body slightly shining, remainder of body, including the frontal area, opaque.

Head, thorax, petiole, and base of gaster with short, rather dense hair; pubescence grayish, moderately developed on these and the remaining portions of the body and appendages. Eyes distinctly hairy.

Deep black even to the tips of the mandibles and appendages; genitalia yellowish, the separate sclerites tipped and bordered with black and castaneous. Wings grayish hyaline, distinctly infuscated towards their bases; veins dark brown, stigma black.

HOST (TEMPORARY). Unknown; probably *F. fusca* var. *argentea.*

TYPE LOCALITY.— Colorado (Mayr).

Colorado: Manitou, Ute Pass, Colorado City, Colorado Springs, Malvern, Wild Horse (7,000–8,000 ft.) (Wheeler).

Montana: Elkhorn (W. M. Mann).

The aberrant type of female, with its remarkable pilosity so much like the trichomes of many myrmecophilous beetles, suggests that this ant must be a temporary parasite on some one of the Colorado varieties of *F. fusca,* but up to the present time it has not been taken in mixed colonies. Although the female may be distinguished at a glance from the females of any of the known species of Formica, the worker and male are not so easily recognized, since they closely resemble the various western forms of *rufa* and *truncicola* and the two following species, *comata* and *criniventris.* The ground color and pilosity of the gaster of the worker are, nevertheless, peculiar, the erect hairs being very short and stubby, and more abundant than in any of the foregoing species.

45. F. COMATA Wheeler.

F. comata Wheeler, Journ. N. Y. ent. soc., 1909, **17**, p. 85, ☿ ♀ ♂.

WORKER. Length 4.5–7 mm.

Allied to *F. ciliata* Mayr. Head, excluding the mandibles, as broad as long, broader behind than in front, with rounded posterior corners, feebly excavated posterior margin, and slightly convex sides. Eyes large. Mandibles 8-toothed. Clypeus carinate, with broadly rounded, entire anterior border, not projecting in the middle. Frontal area subsemicircular, broader than long. Antennal scapes straight at the base, slightly enlarging distally; funicular joints 1–4 somewhat more slender than the remaining joints. Palpi short. Thorax as usual in the *rufa* group of Formica, epinotum angular in profile, with subequal base and declivity, the former horizontal and slightly convex, the latter sloping and slightly concave. Petiole as high as the epinotum, in profile attenuated above, with rather sharp border; seen from behind rounded or more often produced upward in the middle in the form of a blunt point; anterior surface convex, posterior surface flat. Gaster rather large, legs of the usual configuration.

Subopaque, slightly glossy; corners of head somewhat shining; whole body finely and densely shagreened; frontal area, bases of mandibles and corners of clypeus glabrous; mandibles finely and densely striated.

Hairs yellow, short and suberect, sparse on the head, thorax, and petiole, more abundant and obtuse on the gaster, absent on the antennal scapes, present on the gula and in a single row on the flexor surfaces of the femora and tibiae, scattered on the fore coxae, long on the venter, and tip of gaster. Eyes hairless. Pubescence long, grayish, sparse on the head, thorax, and petiole, dense on the gaster, where it completely conceals the surface; somewhat conspicuous on the legs.

Yellowish red; gaster blackish brown, except a large spot at the base and the anal region, which are reddish or yellowish. Mandibles, corners of clypeus, antennae, and legs reddish brown; bases of scapes often paler; pro- and mesonotum each with a fuscous spot, pale or absent in the largest, somewhat larger and darker in the smallest workers; apical half of petiolar node more or less infuscated. Small workers also with brown or black spots on the clypeus, front, occiput, and epinotum and with the coxae more or less infuscated. Mandibular teeth black.

FEMALE. Length 7.5–8 mm.

Resembling the female *ciliata* in form. Whole body much more shining than that of the worker as the shagreening of its surface is much more delicate; scutellum and metanotum glabrous. Pubescence like that of the worker, but longer; pilosity grayish, resembling that of the female *ciliata* but less dense, and the very long hairs on the gaster are slender, less appressed, rather straight, and not recurved at their tips. Color of the body dull brownish yellow, gaster blackish brown, except its base and anal region. Mandibles, funiculi, corners of clypeus, anterior borders of cheeks, posterior border of pronotum, a large anteromedian and two parapsidal blotches on the mesonotum, dull brown; scutellum and metanotum chestnut-brown. Wings long (9 mm.), uniformly infuscated, with brown veins and darker stigma.

MALE. Length 8–8.5 mm.

Head decidedly broader than long, narrowed in the region of the cheeks, which are short and flat; posterior border of head straight, posterior corners broadly rounded. Eyes large, suboblong. Maxillary palpi 5-jointed. Mandibles 4-toothed. Clypeus convex, subcarinate, with entire, slightly reflected anterior border. Thorax and gaster of the usual shape, the former distinctly broader than the head. Petiole broad and low, with thick, rounded, transverse upper border.

Body subopaque; pleurae, scutellum, metanotum, and gaster more shining. Mandibles striatopunctate. Head and thorax very finely and densely punctate, gaster shagreened, with rather coarse, scattered piligerous punctures on its upper surface.

Hairs and pubescence grayish, more abundant than in the worker; the hairs very long on the epinotum, petiolar border, basal gastric segment and venter; somewhat shorter on the clypeus and pronotum and still shorter on the upper surface of the gaster. Eyes hairless.

Black; borders of mandibles, tibiae, tarsi, and articulations of legs brownish, or in some specimens yellowish. Genitalia sordid yellow, the tips of the appendages not infuscated.

HOST. Unknown; probably *F. fusca* var. *argentea* or var. *neoclara.*

TYPE LOCALITY.— Colorado: Manitou (Wheeler).

Colorado: Red Rock Canyon, near Colorado City (Wheeler).

South Dakota: Harding County (S. S. Visher).

The female *F. comata,* though it superficially resembles several of the foregoing species, is nevertheless very distinct in sculpture, color, and pilosity. It is much more difficult to distinguish the worker, as it is extremely like the corresponding phase of *F. ciliata,* differing only in having a somewhat more hairy body, darker gaster and in large specimens in having the petiole narrower and with its superior border more pointed and produced upward. The worker *obscuripes* and *aggerans* have more abundant and more erect hairs on the thorax and the infuscation of workers of all sizes is much more pronounced and extensive. The male *comata* is distinguished from the male *ciliata* by its quadridentate mandibles, pale genitalia, and somewhat paler wings.

The nests of *comata* are very similar to those of *ciliata,* being under clusters of stones or about stumps and logs. These objects are rather heavily banked or even covered with vegetable detritus. The winged phases were taken July 26 and August 14.

46. F. CRINIVENTRIS Wheeler.

F. crinita Wheeler, Journ. N. Y. ent. soc., 1909, **17**, p. 87, ☿ ♀.
F. criniventris, nom. nov. Wheeler, Psyche, 1912, **19**, p. 90.

WORKER. Length 4–6.5 mm.

Resembling the worker of the preceding species but averaging somewhat smaller. Head, excluding the mandibles, a little longer than broad, even in the largest workers; narrower in front than behind with nearly straight posterior and lateral margins. Eyes rather large. Mandibles 7–8 toothed. Clypeus carinate, with entire anterior border, slightly projecting in the middle. Frontal furrow distinct. Antennae, thorax, and petiole as in *comata.* Palpi rather short. Gaster and legs of the usual shape.

Body subopaque, very finely shagreened; bases of mandibles, frontal area, and corners of clypeus glabrous. Mandibles and clypeus finely, longitudinally striated.

Hairs yellow; absent on the head, thorax, petiole, and appendages, blunt and scattered on the gaster, pointed on the clypeus, mandibles, and venter. Pubescence yellowish and very short, inconspicuous on the head, thorax, and petiole, somewhat longer on the legs and gaster; on the latter rather dense and nearly concealing the surface. Eyes hairless.

Yellowish red; gaster dark reddish brown, except the anal region and a spot at the base of the first segment, which are yellowish; tips of antennal funiculi, middle portions of femora and tibiae brownish or reddish. The smallest workers have the upper surface of the thorax, especially the pro- and mesonotum, somewhat infuscated. Mandibular teeth black.

FEMALE. Length 6.5–7 mm.

Resembling the female of *ciliata.* Body shining throughout, very finely shagreened, without pubescence. Hairs very long, yellow, curled or hooked at their tips, confined to the clypeus, gaster, and ventral surface of the petiole; on the gaster appressed and arranged in two rows near the posterior border of each segment. Body and appendages yellow; teeth of mandibles and anterior edge of clypeus black; scutellum, metanotum, and anteromedian and two parapsidal blotches on the mesonotum, anterior borders of cheeks, and a narrow band parallel with the posterior edge of each gastric segment, brown. Antennal funiculi infuscated towards their tips. Wings grayish hyaline, with pale brown veins and darker brown stigma.

HOST. Unknown, probably *F. fusca* var. *argentea* or *neoclara.*

TYPE LOCALITY.— Colorado: Boulder, (Wheeler).

Montana: Helena (Hubbard and Schwarz).

South Dakota: Harding County (S. S. Visher).

The worker differs from those of *ciliata, comata,* and *oreas* in the absence of hairs on the head, thorax, and petiole, and the female has much fewer hairs and these are confined to the clypeus and gaster. The hairs are very easily rubbed off in both workers and females, but the long series of the former and the callows of the latter show that they cannot be more abundant than described above. The colony from which the type specimens were taken was very populous. Its nest resembled very closely those of *ciliata, comata,* and *oreas* which I have examined in Colorado. It was under several contiguous stones, banked with vegetable detritus and in the immediate neighborhood of flourishing colonies of *F. ciliata* and *rufa aggerans.*

47. F. OREAS Wheeler.

F. oreas Wheeler, Bull. Amer. mus. nat. hist., 1903, **19**, p. 643, ☿ ♀ ♂.

WORKER. Length 4.5–7 mm.

Resembling the workers of *F. ciliata, comata,* and *criniventris.* Mandibles 8-toothed. Head, excluding the mandibles, as broad as long, slightly narrower in front than behind, with feebly concave posterior border, rather broadly rounded posterior corners, and convex sides. Clypeus carinate its entire length, with broadly rounded, not produced anterior border. Antennae rather slender, funicular joints 1–3 longer and more slender than the penultimate joints. Frontal carinae rather strongly diverging. Palpi short. Thorax with convex pro- and mesonotum and deep mesoëpinotal constriction. Epinotum with the base horizontal and slightly convex, distinctly longer than the rapidly sloping and distinctly concave declivity with which it forms an obtuse angle. Petiole broad, compressed anteroposteriorly with a sharp superior border, which is either bluntly pointed or slightly truncated in the middle.

Body subopaque, very finely shagreened, anterior portion of head smooth and shining, mandibles and clypeus longitudinally striated, shining; frontal area glabrous.

Hairs silvery white or pale yellow, short, abundant, erect, covering both the dorsal and gular surfaces of the head, the thorax, border of petiole, and gaster. Hairs on the ocellar region conspicuously long. Scapes and legs covered with shorter, suberect hairs. Hairs on the gaster pointed and more delicate than in the three preceding species, long on the venter and terminal segments. Eyes hairy. Pubescence yellowish, sparse on the head, somewhat more abundant on the thorax and sufficiently dense on the gaster to conceal the surface and give it a grayish tinge.

Bright yellowish red, mandibles and antennal scapes darker; funiculi and legs reddish brown, their articulations more yellowish. Gaster black, anal segment, a large spot at the base of the first segment and often a spot on each of the sternites, yellow or red. Some of the smallest workers have the vertex, pro- and mesonotum blotched with black, but others have the head and thorax almost as immaculate as large individuals.

FEMALE. Length 7.5–9 mm.

Head resembling that of the worker but with less convex sides. Carina of clypeus delicate. Thorax nearly as broad as the head, robust. Petiole very broad, much compressed anteroposteriorly, feebly convex in front, flattened behind, the sharp border broadly rounded, more rarely bluntly angular in the middle.

Mandibles striatopunctate, somewhat less shining than the remainder of the body, which, together with the appendages, is smooth; the upper surface of the head, mesonotum, and scutellum, especially are highly glabrous.

Entire body, including the antennal scapes and legs, covered with short, delicate, erect or suberect, silvery white hairs which nowhere conceal the shining surface. These hairs are abundant and conspicuous on the scapes, legs, and gaster, less abundant on the front of the head, and on the mesonotum, at least in some specimens. Eyes hairy.

Color rich yellowish red; mandibular teeth, anterolateral borders of clypeus, thoracic sutures, alar insertions, metanotum, and adjacent border of scutellum, posterior border of each gastric segment, palpi, articulations of legs, and terminal half of funiculi, black or infuscated. Wings uniformly gray, veins and stigma sordid yellow.

MALE. Length 7–8 mm.

Mandibles 3–4-toothed. Maxillary palpi slender, 5-jointed. Head small, eyes large, cheeks rather concave, postocular region less prominent than in the male *ciliata*. Clypeus sharply carinate. Thorax robust, decidedly broader than the head. Petiole low, cuneate in profile, thick at base, but becoming rapidly thin towards the superior border, which when seen from behind is feebly and broadly excised in the middle.

Body opaque, finely shagreened; frontal area shining; mesonotum, scutellum, and gaster above slightly lustrous.

Hairs gray, dense, longest and suberect on the head, thorax, and petiole, more reclinate on the gaster, very short and subappressed on the legs and antennae. Eyes distinctly hairy. Gaster delicately gray pubescent.

Deep black; genitalia, tips of trochanters, knees, basal portions of first and second tarsal joints, reddish yellow. Wings like those of the female, except that the stigma is darker.

HOST. Unknown; probably one of the subalpine varieties of *F. fusca.*

TYPE LOCALITY.— Colorado: Woodland Park, Ute Pass, 8,500 ft. (Wheeler).

Colorado: Buena Vista, Boulder, Wild Horse, Salida, Florissant (Wheeler).

New Mexico: Embudo (T. D. A. Cockerell).

The female of this species is readily distinguished from the female of *criniventris* and *ciliata* by its pilosity and from the females of all the other species described above by its color and the erect hairs on the antennal scapes. This last character also enables one to separate the worker from the very similar workers of all the foregoing species of the *rufa* group.

There can be little doubt that this ant is a temporary parasite on some form of *F. fusca.* The nests which I saw in the localities recorded above during the summer of 1903 and 1906 were not abundant but were very populous. They were established in open, sunny places, under stones, the edges of which were heavily banked with vegetable detritus.

48. F. OREAS Wheeler var. COMPTULA, var. nov.

WORKER. Length 3–7 mm.

Differing from the worker of the typical *oreas* in color and pilosity. The red portions of the body are darker and less yellowish, the gaster blacker, the legs dark brown, or nearly black, with red articulations. In small workers the vertex, upper surface of thorax and petiole are rather heavily infuscated. The erect hairs on all parts of the body, especially on the head, are somewhat more abundant; the hairs on the gaster though very numerous are only half as long as in *oreas,* and as the pubescence is shorter and sparser on this region, it appears blacker and less glaucous.

FEMALE (DEÄLATED). 7.5–8 mm.

Differing from the female of the typical *oreas* in having the white hairs covering the body, scapes, and legs conspicuously more abundant and somewhat coarser. On the legs and scapes the hairs are more erect, and they are very dense on the epinotum, petiole, and upper surface of the gaster.

Described from two females and ten workers taken by Mr. Wm. M. Mann from a large colony at Pullman, Washington. Mr. Mann has also taken workers and females of this variety at Elkhorn, Montana.

49. F. FEROCULA, sp. nov.

WORKER. Length 3.5–6 mm.

Head, excluding the mandibles, as broad as long, broader behind than in front, with feebly excised posterior border and very slightly convex sides. Mandibles 8-toothed. Clypeus convex, carinate its entire length, with broadly rounded anterior border, but slightly or not at all produced in the middle. Frontal area triangular, as long as broad. Frontal carinae short, diverging. Antennae slender, four basal funicular joints longer and more slender than the penultimate joints. Palpi short. Pro- and mesonotum not very convex, mesoëpinotal constriction not very deep, epinotum with subequal base and declivity, the former feebly convex, the latter distinctly concave. Petiole narrow and very low, its anterior surface very convex, its posterior surface flattened, its border very blunt and when seen from behind, evenly rounded and entire, not produced upward in the middle. Legs rather long.

Opaque, finely shagreened; mandibles shining, rather superficially striatopunctate; frontal area smooth and shining, clypeus also more shining than the posterior part of the head.

Hairs and pubescence golden yellow; the former abundant on the clypeus and mandibles, absent on the remaining portions of the head; dense and erect on the pronotum, epinotum, and petiole, absent on the mesonotum, except at the posterior border. On the gaster the erect hairs are short, obtuse, and rather abundant. Eyes hairless. Pubescence long and rather dense on the head and thorax, scarcely denser on the gaster and not concealing the ground color. Pubescence on the legs long and somewhat oblique on the flexor surfaces of the tibiae.

Bright yellowish red; mandibles, antennal funiculi towards their tips, and the legs in some specimens, darker. Gaster dark brown, with the anal region and a spot at the base of the first segment red. Very small workers have the upper surface of the head, thorax, and petiole infuscated and the legs darker.

Described from sixteen workers taken from a single colony at Rockford, Illinois. This and several other colonies of the same species were found nesting in dry, open fields in crater nests 3–4 inches in diameter about the roots of *Erigeron canadense* and other weeds. The species is evidently allied to *F. comata, criniventris, ciliata,* and *oreas,* but differs from all of these forms in the peculiar shape of the petiole and the arrangement of the hairs. The female is probably of a peculiar aberrant type, like the females of the forms just mentioned.

50. F. DAKOTENSIS Emery.

F. dakotensis Emery, Zool. jahrb. Syst., 1893, **7**, p. 652, taf. 22, fig. 5, ☿.
F. subpolita var. ? *specularis* Emery, Zool. jahrb. Syst., 1893, **7**, p. 663, ♀ (*in part*).

WORKER. Length 5–6 mm.

Mandibles 8-toothed. Head, excluding the mandibles, as broad as long, a little broader behind than in front, with feebly excised posterior margin, broadly rounded posterior corners, and convex sides. Clypeus convex, sharply carinate, its anterior border not produced, broadly rounded. Frontal area triangular, as long as broad; frontal carinae strongly diverging. Antennal scapes slender at the base, distinctly enlarged at their tips; funicular joints 2–4 a little more slender and not much longer than the penultimate joints. Palpi very short. Thorax shaped much like that of *F. sanguinea,* with moderately convex pro- and mesonotum, moderately deep mesoëpinotal constriction and the epinotum rather angular, with subequal base and declivity, the former straight and horizontal in profile, the latter slightly concave. Petiole narrow, in profile cuneate, thick below, with rather strongly convex anterior, more feebly convex posterior surface, and blunt superior margin. Seen from behind it is narrow below, but gradually broadening dorsally, with straight sides and horizontal or feebly and broadly excised superior border, so that it appears truncated above.

Subopaque, very delicately shagreened; mandibles and clypeus more shining, the former finely striatopunctate; clypeus very finely longitudinally striated. Frontal area smooth and shining. Gaster more distinctly shagreened than the body, shining with the same luster as in *F. truncicola obscuriventris* and *F. exsectoides.*

Upper surface of head, thorax, and petiole without hairs or with a very few scattered yellow hairs. Those on the gaster longer, blunt and scattered, apparently deciduous on the upper surface, longer and more abundant on the venter. Gula and eyes hairless. Pubescence very fine and sparse, scarcely perceptible on the body, more distinct on the legs and scapes.

Light or dark red, funiculi darker, gaster black or very dark brown, the base of the first segment not distinctly reddish.

FEMALE (After Emery). Length 7.5–8 mm.

Antennae, metanotum, tibiae, tarsi, and whole of gaster brown, first gastric segment ferruginous red at the base. The whole insect very shining; mesonotum and gaster as smooth as a mirror, the latter with only a few small punctures which bear the very sparse, short pubescence; erect hairs rather numerous on the base, tip, and venter of the gaster, sparse and very short on the head, thorax, and petiole; lower surface of head without hairs. Pubescence on head and thorax dilute and very short, completely lacking on the mesonotum. Head broad behind and with a straight margin and rounded posterior corners. Clypeus broadly rounded, scarcely carinate, shining, feebly and obliquely striated. Mandibles shining, strongly sculptured; petiole cuneate, squarely truncated above.

HOST (TEMPORARY). Probably *F. fusca* var. *subsericea*.

TYPE LOCALITY.— South Dakota: Hill City (Th. Pergande).

British Columbia: Golden (W. Wenman).

Nova Scotia: Digby (J. Russell).

I have redescribed the worker of this species from a cotype. It is easily recognized by the shape of the head and especially by the petiole, which differs from that of all other species of Formica known to me. The palpi, too, as Emery has observed, are remarkably short. The specimens from the three localities mentioned above all agree in having extremely few or no hairs on the dorsal surface of the body and none on the gula, thus coinciding with Emery's remark "superne haud pilosa," so that the following form, which I first described as a distinct species and later regarded as identical with the typical *dakotensis*, may be retained as a variety.

51. F. DAKOTENSIS var. MONTIGENA Wheeler.

F. montigena Wheeler, Bull. Amer. mus. nat. hist., 1904, **20**, p. 374, ☿ ♀ ♂.

WORKER. Length 3.5–6.5 mm.

Differing from the worker of the typical *dakotensis* in having longer and more numerous erect hairs on the upper surface of the head, thorax, and petiole, and in having a few erect hairs on the gula. The pubescence on the gaster and legs seems also to be a little longer and more distinct. The gaster is more brownish or reddish and the base of the first segment is often yellow or red.

FEMALE. Length 7 mm.

Mandibles and clypeus like those of the worker. Head large, as broad as long, its sides straight, slightly converging in front, its posterior angles rounded, its posterior border feebly excised. Thorax distinctly narrower than the head. Petiole extremely thick and blunt, its upper border transverse and feebly excised when seen from behind. Wings as long as the body (7 mm.).

Body and legs very glabrous and shining. Mandibles coarsely striatopunctate. Clypeus delicately striated anteriorly. Antennae subopaque.

Hairs suberect, sparse, yellowish, longest on the gaster, especially towards its tip, shorter on the head and thorax and confined to the flexor surfaces of the femora and tibiae. Pubescence grayish, very dilute and inconspicuous, except on the antennae.

Rich yellowish red. Mandibles, corners of clypeus, tarsi, and antennal scapes darker. Mandibular teeth black; funiculi, gaster, scutellum, metanotum, a triangular anterior, and two elongate parapsidal blotches on the mesonotum dark brown. Wings grayish hyaline, not infuscated, with pale brown veins and darker stigma.

MALE. Length 6.5–7 mm.

Mandibles 3–4 toothed. Head very short and broad, narrow in front, posterior corners prominent and rounded; eyes large; cheeks short, concave. Clypeus convex, carinate. Thorax broad and robust. Petiole low, thick, transverse, with very blunt border, which, seen from behind, is broadly and distinctly excised.

Body subopaque; mandibles and frontal area slightly, upper surface of gaster more distinctly shining.

Hairs and pubescence sordid yellow, sparse, and inconspicuous, especially on the upper surface of the gaster.

Black. Genitalia reddish yellow. Legs sordid yellow; femora, especially the fore pair, more or less infuscated. Wings whitish hyaline, with pale brown veins and slightly darker stigma.

HOSTS (TEMPORARY). *F. fusca* var. *subsericea*, and *F. pallidefulva schaufussi* var. *incerta*.

TYPE LOCALITY.— Colorado: Ute Pass and Pike's Peak, 10,000–11,500 ft. (Cockerell and Wheeler).

Colorado: North Cheyenne Canyon, near Colorado Springs, Florissant, Buena Vista (Wheeler).

New Mexico: Beulah, 8,000 ft. (T. D. A. Cockerell).

Montana: Helena (W. M. Mann).

Idaho: Troy (W. M. Mann).

That this variety is a typical temporary parasite is shown by the fact that I found two small mixed colonies (with *F. incerta*) on Pike's Peak and that Mr. Mann found a single colony mixed with *F. subsericea* at Troy, Idaho. The adult colonies are large, and form nests under stones or about the roots of plants which are banked with considerable vegetable detritus.

52. F. DAKOTENSIS var. SPECULARIS Emery.

F. subpolita var. ? *specularis* Emery, Zool. jahrb. Syst., 1893, **7**, p. 663, ♀ (*in part*).

F. dakotensis Wasmann, Allgem. zeitschr. ent., 1901, **6**, p. 6.

F. dakotensis Emery var. *wasmanni* Forel, Ann. Soc. ent. Belg. 1904, **48**, p. 153, *nota* ☿ ♀ ♂.

WORKER. Length 5–7 mm.

Differing from the typical *dakotensis* in having the sides of the head a little less convex, the anterior border of head a little narrower, and the gaster somewhat paler. The pubescence is longer and more distinct on head and thorax. The pilosity is that of the type and not that of the var. *montigena.*

FEMALE. Length 7–8 mm.

Differing from the females of the typical *dakotensis* and the var. *montigena* in color, being yellowish red except the posterior borders of the gastric segments, the antennae, and tibiae which are brown, and the mandibles which are deep red. Wings with a yellowish tinge, veins light brown, stigma darker.

MALE. Length 7.5–8 mm.

Closely resembling the male of the var. *montigena,* but the legs and genitalia of a purer yellow and the gaster dark brown instead of black. This is the case in ten specimens in my collection and can hardly be due to immaturity. Wings colored as in the female.

HOST (TEMPORARY). *F. fusca* var. *subsericea.*

TYPE LOCALITY.— Wisconsin (Wasmann).

Wisconsin: Prairie du Chien (H. Muckermann).

Emery described two different females under the name *specularis,* the first belonging to this variety, the second to the typical *dakotensis.* Muckermann found several colonies of *specularis* mixed with *subsericea* and Wasmann concluded from this that the former ant was an incipient slave-maker. It is evident that this conclusion is erroneous, since my observations on the var. *montigena,* show that the species is a temporary social parasite, like many, if not all, other forms of the *rufa* group.

Microgyna Group.

53. F. MICROGYNA MICROGYNA Wheeler.

F. microgyna Wheeler, Bull. Amer. mus. nat. hist., 1903, **19**, p. 645, fig. 3, ☿ ♀ ♂, p. 656, fig. 1 (gynandromorph).

WORKER. Length 3.5–6 mm.

Mandibles 8-toothed. Clypeus rounded in front, not produced, carinate its entire length and with uneven surface. Maxillary palpi rather long. Head, excluding the mandibles, somewhat longer than broad even in the largest workers, but little narrower in front than behind, with straight posterior border and straight subparallel sides.

Antennae rather slender, scapes but slightly enlarged toward their tips, joints 2–4 of the funiculus decidedly longer and more slender than the penultimate joints. Frontal area semicircular, broader than long, frontal carinae distinctly diverging behind. Pro- and mesonotum convex, mesoëpinotal constriction narrow and rather deep, epinotum rounded, its base convex. Petiole narrow, its anterior surface convex, its posterior surface more flattened, with entire, blunt border, projecting upward in the middle in some specimens, in others evenly rounded or even slightly excised.

Body opaque, finely shagreened. Mandibles opaque and finely striated apically, smooth and shining at the base. Clypeus opaque, very finely striated. Frontal area smooth and shining.

Entire insect, including appendages, covered with microscopic gray pubescence, dense and most distinct on the gaster. Hairs pale yellow, erect, and, except on the mandibles, distinctly clavate, with obtuse tips. These hairs are sparse on the clypeus, on the front, where they form four longitudinal rows as far back as the ocelli, on the thoracic dorsum, coxae, border of the petiole, and surface of the gaster. On the last they are particularly conspicuous on account of their equidistant arrangement and contrasting color. They are easily rubbed off. A few hairs are occasionally present on the gula but usually entirely absent. Anterior border of scape with a row of delicate, suberect, tapering hairs. Tibiae covered with very short, stiff, suberect hairs. Eyes hairless.

Head, thorax, and petiole deep yellowish red, mandibles and clypeus somewhat darker, ocellar region often fuscous. In small workers the front, vertex, thoracic dorsum, and petiole are spotted with black. Gaster black, with only the anal region yellowish. In the largest workers the legs are red throughout, in intermediates the femora and tibiae are brownish, in the smallest workers the infuscation extends also to the coxae. Antennae red, funiculi more or less infuscated toward the tip.

Female. Length 4–4.5 mm.

Head resembling that of the worker. Thorax distinctly narrower than the head. Petiole narrow, thick at the base, both its anterior and posterior surfaces alike convex, its dorsal border sharper than in the worker; seen from behind variable, in some specimens evenly rounded, in others somewhat produced upward in the middle.

Sculpture like that of the worker. Whole insect, including the antennae and legs, clothed with delicate white hairs, which are longer and more abundant than in the worker and not clavate though obtuse. These hairs are conspicuously long and suberect on the front, gula, thorax, petiolar border, gaster, antennal scapes, and legs. In addition to these hairs the body and appendages are invested with microscopic, white pubescence.

Head, thorax, petiole, and legs dull, reddish or brownish yellow. Mandibular teeth, funiculi, a blotch covering the ocellar region, a large anteromedian mesonotal, and two elongate parapsidal blotches, alar insertions, metanotum, and more or less of the contiguous portion of the scutellum, fuscous. In some specimens the clypeus, frontal region, coxae, and pleurae are infuscated. Gaster dark brown, anal segment, and more or less of the base of the first gastric segment, brownish yellow. Wings whitish hyaline, veins and stigma brown, the latter conspicuous.

MALE. Length 5–5.5 mm.

Mandibles slender, edentulous or indistinctly 3-toothed, pointed. Maxillary palpi 5-jointed. Head rather short, broad, and convex behind the eyes, narrow in the region of the cheeks. Eyes large. Clypeus distinctly carinate anteriorly. Antennae slender. Thorax broader than the head, rather robust, mesonotum flattened in front of the scutellum. Petiole very thick, with obtuse and broadly rounded border. Genitalia rather slender.

Subopaque; frontal area, anteromedian suture of mesonotum, parapsidal furrows, paraptera, and upper surface of gaster smooth and shining. Mandibles coarsely punctate near the tips, finely striated toward the base. Frontal area slightly shining.

Body and appendages clothed with microscopic grayish pubescence, which is sparse and visible only in certain lights. Hairs covering the body and appendages delicate, sparse, suberect, of an indistinct grayish color. Eyes naked.

Deep black: legs and genitalia dirty yellow; coxae, femora, tibiae, and terminal tarsal joints more or less infuscated. Wings like those of the female.

HOSTS (TEMPORARY). *F. fusca* var. *argentea* and *F. neogagates.*

TYPE LOCALITY.— Colorado: Manitou (Wheeler).

Colorado: Florissant (Wheeler).

The colonies of this and the following species, here included in the *microgyna* group, are much smaller than those of the *rufa* group, but the nests are much like those of *truncicola* and the allied forms, being under single stones or clusters of stones, which are banked with more or less vegetable detritus.

54. F. MICROGYNA MICROGYNA var. RECIDIVA, var. nov.

WORKER. Length 3.5–6 mm.

Differing from the worker of the typical form in lacking the erect hairs on the antennal scapes and in having the hairs on the tibiae shorter, more abundant, and somewhat more appressed.

MALE. Indistinguishable from the male of the typical *microgyna.* Mandibles indistinctly toothed in two specimens.

Described from two males and numerous workers taken from three colonies at Florissant, Colo. Four workers taken by Prof. T. D. A. Cockerell at Pecos, New Mexico are also referable to this variety. One occasionally finds workers which are intermediate between this variety and the type in possessing a few erect hairs on the anterior surface of the antennal scapes near the base. The nests are in all respects like those of the typical form.

55. F. MICROGYNA RASILIS Wheeler.

F. microgyna var. *rasilis* Wheeler, Bull. Amer. mus. nat. hist., 1903, **19**, p. 648, ☿ ♀ ♂.

WORKER. Length 4–6.5 mm.

Closely related to the typical *microgyna* but averaging a little larger. Head slightly longer than broad, very nearly as broad in front as behind, with feebly concave posterior border in the largest individuals and nearly straight sides. Clypeus convex, carinate its entire length, its anterior border somewhat angularly projecting in the middle. Frontal area triangular, as long as broad. Frontal carinae diverging. Antennae rather slender, the basal joints much longer and more slender than the penultimate joints. Maxillary palpi rather long. Thorax shaped as in *microgyna,* the petiole with a somewhat sharper border.

Sculpture as in *microgyna.* Mandibles and frontal area but slightly shining. Legs rather smooth.

Hairs golden yellow, obtuse, sparser than in *microgyna,* and stouter, absent on the antennal scapes, gula, and posterior corners of the head, and the eyes and legs, except for the row of bristles on the flexor surfaces of the tibiae, entirely naked. Hairs on the clypeus, thorax, and petiole much less numerous. Pubescence very fine and uniform, not very dense on any part of the body. Color like that of *microgyna.*

FEMALE. Length 5–5.5 mm.

Averaging a little larger than the female of *microgyna* and differing in pilosity, sculpture, and color. The gaster is more opaque, the black blotches on the head and thorax are indistinct or entirely lacking, even in mature specimens. The wings are more grayish and the hairs on the head, thorax, petiole, and gaster are sparser, stouter, more clavate and more obtuse. The antennal scapes and legs are without erect hairs.

MALE. Length 6–7 mm.

Mandibles edentate or quadridentate, their blades broader than in *microgyna.* Frontal area smooth and shining. Erect hairs on gula less abundant as are also the oblique hairs on the legs. Wings somewhat more opaque.

HOST (TEMPORARY). *F. fusca* var. *argentea* and var. *subsericea.*

TYPE LOCALITY.— Colorado: Manitou (Wheeler).

Colorado: Ute Pass, Pike's Peak (11,500 ft.), Colorado Springs, Florissant, Wild Horse (Wheeler).

New Mexico: Pecos (T. D. A. Cockerell).

Utah: Salt Lake (R. V. Chamberlin).

Washington: Olympia (T. Kincaid).

This form was originally described as a variety of *microgyna* but after examining more material than I possessed at the time of its description, I am convinced that it should have subspecific rank, at least. It has apparently the same range as the typical *microgyna*, but is more abundant and forms more populous colonies. These live under stones and may occupy several nests covering an area of a square meter or more. Between July 11 and August 21, 1903, I found, in all, thirteen of these colonies in the neighborhood of Manitou, Colo. and during July 1906 I found nearly as many more at Florissant, and Wild Horse in the same state. A few of these colonies were very small and young and mixed with workers of *F. fusca* var. *argentea* and var. *subsericea,* proving that *rasilis* is a temporary social parasite on these ants.

56. F. MICROGYNA RASILIS var. SPICATA, subsp. nov.

WORKER. Length 3.5–6 mm.

Closely resembling the worker of *rasilis* but even the largest individuals have the pro- and mesonotum and nearly always also the ocellar region infuscated or blackened, and the hairs on the gaster are longer and slightly more numerous. The pubescence on the gaster is denser so that this region is gray and its ground color is concealed. Legs and antennal scapes without erect hairs. Gula in some specimens pilose. Frontal area very smooth and shining.

FEMALE. Length 4.5–5 mm.

Differing from the female *rasilis* in having the blunt clavate hairs on the head, thorax, and gaster longer and more numerous. This is especially true of those on the gaster. Gaster opaque, owing to the dense and rather long pubescence. Body and legs brownish yellow, with the upper surface of the head, mesonotum, scutellum, meso- and metapleurae, gaster, antennal funiculi, and usually also the coxae dark brown. Wings grayish hyaline.

MALE. Length 5.5–6 mm.

Mandibles broad, usually edentate, more rarely obscurely dentate. Petiole very low, scarcely higher than long, its upper surface in profile obliquely flattened above so that the posterior surface is higher than the anterior. Seen from behind the upper border is entire and broadly rounded. In *rasilis* and the typical *microgyna* the border is sharper and more compressed. Frontal area very smooth and shining. Color, sculpture, and pilosity as in these forms. Upper surface of head and thorax, and the gula with rather numerous erect hairs; tibiae with numerous oblique hairs. Wings as in the female.

Described from numerous workers, males, and rather immature females taken from five colonies at Florissant, Colorado (altitude 8,100 ft.).

57. F. MICROGYNA SCITULA, subsp. nov.

FEMALE. Length 4.5 mm.

Closely resembling the female of *rasilis* in color, except that the base of the first gastric segment is brownish red like the head; thorax, petiole, legs, scapes and base of the funiculi, and the wings are faintly but distinctly infuscated. Anal region reddish. Gaster and terminal funicular joints dark brown. The clavate hairs on the head, thorax, and gaster are as long as in the var. *spicata* but more numerous on the posterior portion of the pronotum and the whole of the mesonotum. Gula with a very few erect hairs. Pubescence very fine and indistinct, except on the gaster. There are no oblique or suberect hairs on the scapes or legs. The body, including the legs and gaster, is smooth and slightly shining, the frontal area subopaque.

Described from a single specimen taken during June 1909 by Mr. W. T. Davis at Clayton, Georgia, 2,000–3,700 ft.

This specimen is so much like the female *microgyna*, and especially those of its subspecies *rasilis* and the var. *spicata* that I feel compelled to place it here, although the locality is far distant from the range of the allied forms. Its ultimate taxonomic position will, of course, depend on the discovery of the worker and male.

58. F. NEVADENSIS Wheeler.

F. microgyna var. *nevadensis* Wheeler, Bull. Amer. mus. nat. hist., 1904, **20**, p. 373, ♀.

F. nevadensis Wheeler, Bull. Amer. mus. nat. hist., 1905, **21**, p. 272; Ants, 1910, p. 570.

FEMALE. Length 4.5 mm.

Closely resembling the female of the typical *F. microgyna* but differing in the following characters:— The petiole in profile is cuneate,

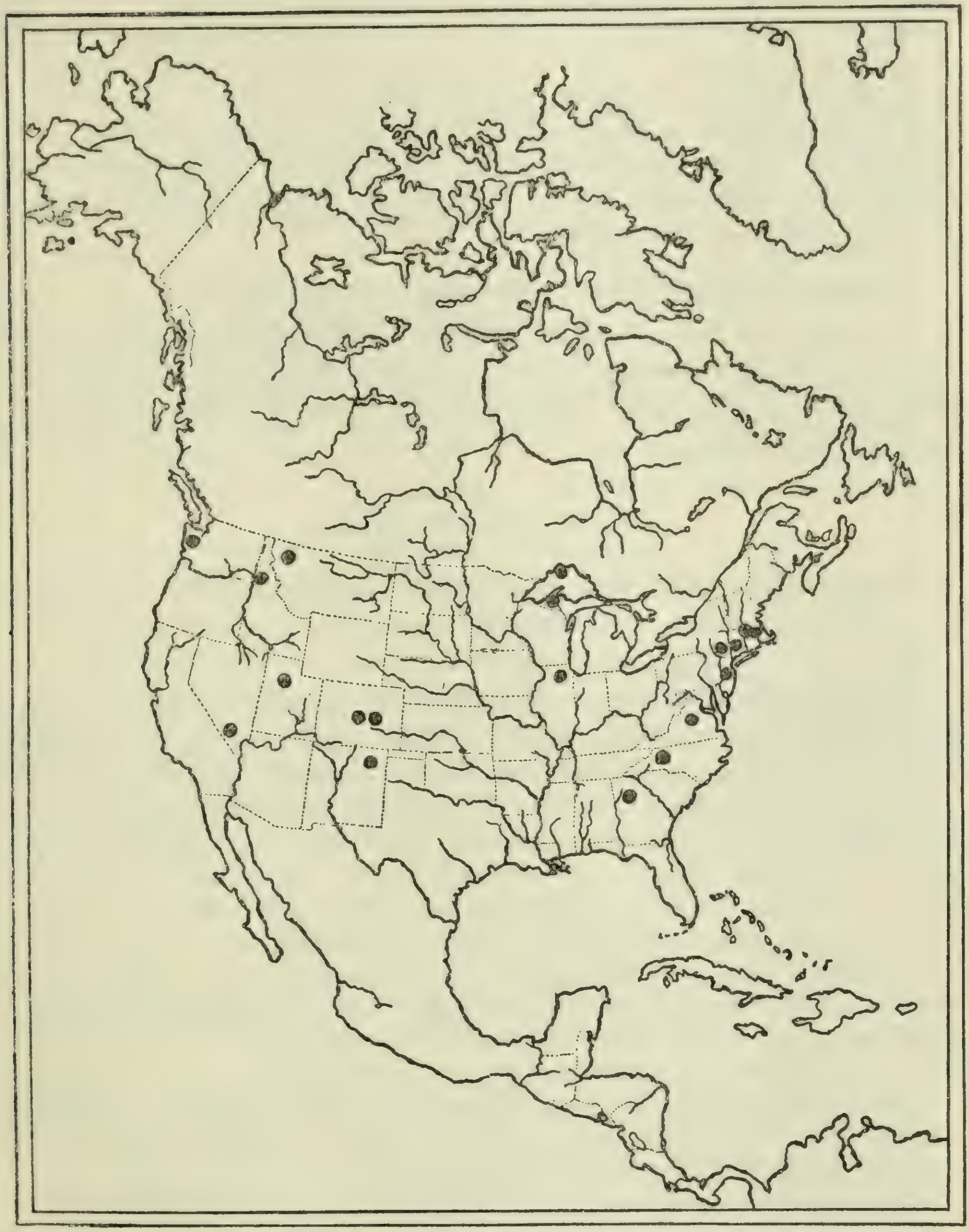

Fig. 4.— Distribution of the forms of the *Formica microgyna* group.

thick below, with very sharp upper border and straight anterior and posterior surfaces. The erect, silvery white hairs covering the body, legs, and antennal scapes are longer and more abundant. The pubescence is very fine and sparse on the gaster so that this region is very smooth and shining. The mesonotum is not spotted but, together with the scutellum, paraptera, and metanotum, uniformly dark brown and rather sharply marked off from the remaining paler portions of the thorax, which are brownish red. Occiput slightly infuscated. Gaster very dark brown, anal region red. Wings grayish hyaline, with brown veins and stigma.

Nevada: Ormsby County, July 1903 (C. F. Baker).

Although this form is readily distinguished from the females of all the forms of *microgyna* by its very smooth gaster, more material may show that it is to be regarded as a subspecies of *microgyna* and not as an independent species.

59. F. IMPEXA Wheeler.

F. impexa Wheeler, Bull. Amer. mus. nat. hist., 1905, **21**, p. 273, ☿; Psyche, 1906, **13**, p. 40, ♀; Ants, 1910, p. 570.

WORKER. Length 3.3–6 mm.

Resembling *F. microgyna*. Mandibles 8-toothed. Clypeus broadly rounded in front, not produced in the middle, carinate its entire length. Head, excluding the mandibles, distinctly longer than broad even in the largest workers, with straight posterior and straight, subparallel lateral borders. Joints 1–4 of funiculi decidedly longer and more slender than the penultimate joints. Maxillary palpi rather short. Thorax with the epinotum low and much rounded, without distinct base and declivity, mesoëpinotal constriction rather narrow and shallow, pro- and mesonotum moderately convex. Petiole narrow, cuneate in profile, its anterior surface convex, its posterior surface flat, its border moderately sharp and produced upward in the middle. In small workers the edge may be much more blunt.

Mandibles lustrous, finely and sharply striated. Surface of clypeus uneven. Frontal area smooth and shining. Remainder of body opaque, finely but distinctly shagreened.

Whole body and appendages clothed with very minute white pubescence, which is rather sparse on the head and thorax, but dense and concealing the ground surface of the gaster. Body, antennal scapes and legs covered with short, coarse, obtuse, erect or suberect, whitish or yellowish hairs. On the gaster these are uniformly distributed and in certain lights very conspicuous, but shorter than in the various forms of *microgyna*. They are also very numerous and prominent on the thoracic dorsum, clypeus, front, vertex, posterior corners and gular surface of head, but absent or very sparse on the cheeks, pleurae, and coxae. They are prominent both on the flexor and extensor surfaces of the legs.

Head and thorax red. Gaster black. Even in the largest specimens the mandibles, anterior border of clypeus and apical half of funiculi are dark reddish brown; ocellar triangle, upper surface of pro- and mesonotum, much of the upper surface of the petiole, legs and coxae, except their articulations, more or less blackened or infuscated. Fore coxae largely red. Anal region yellowish. In the smallest workers the infuscation is more extensive, involving the whole of the posterior portion of the head and epinotum.

Female. Length 4.5 mm.

Resembling the worker in sculpture and pilosity. Head, thorax, petiole, and legs yellowish or reddish brown. Tips of funiculi, scutellum, metanotum, and gaster dark brown; mesonotum with three elongate brown blotches. Wings gray, with brown veins and stigma.

Host (Temporary). *F. fusca* var. *subaenescens.*

Type locality.— Michigan: Porcupine Mts. (O. McCreary).

Massachusetts: East Holliston, Sherborn (A. P. Morse).

This species, too, is very closely related to the typical *F. microgyna,* but both the worker and female are much more densely and coarsely pilose and the epinotum of the worker is peculiarly low and rounded. The colony of this species found by Mr. Morse at Sherborn, Mass. was apparently still mixed with workers of *F. fusca* var. *subaenescens,* although it contained a few winged females of the parasitic species.

60. F. ADAMSI Wheeler.

F. adamsi Wheeler, Journ. N. Y. ent. soc., 1909, **17**, p. 84, ☿; Rept. Mich. geol. survey for 1908, 1909, p. 326 ☿; Ants, 1910, p. 570.

Worker. Length 3.5–5 mm. Allied to *F. microgyna.* Head, excluding the mandibles, a little longer than broad, very nearly as broad in front as behind, with straight sides and straight or slightly concave posterior border. Eyes rather large. Mandibles 7–8 toothed. Clypeus strongly carinate, with broadly rounded anterior border, not produced in the middle. Palpi of moderate length. Antennae slender, scapes nearly straight at the base, funicular joints all distinctly longer than broad, the basal somewhat more slender and longer than the apical joints. Pro- and mesonotum moderately convex, mesoëpinotal constriction rather shallow, broad at the bottom, epinotum with subequal base and declivity, the former slightly convex, the latter flattened or slightly concave; the two surfaces passing into each other through a distinct but rounded angle. Petiole narrow, in profile compressed anteroposteriorly, with convex anterior and flattened posterior surface and sharp superior border, which when seen from behind is rounded and usually but slightly produced upward in the middle.

Opaque throughout, except the bases of the mandibles, the frontal area, and sides of the clypeus, which are shining. Mandibles finely and densely striated. Surface of body densely and indistinctly shagreened.

Hairs and pubescence pale yellow; the latter covering the whole body and appendages, not conspicuous except on the gaster, but even on this region not sufficiently dense to conceal the surface sculpture. Hairs short, sparse and obtuse, in several rows on the gastric segments; on the thorax confined to the upper portions of the pro- and mesonotum, on the head to the clypeus, front, and vertex. The hairs on the mandibles are appressed and pointed, on the palpi short but numerous and conspicuous. Legs and antennae naked, the former only with a series of pointed bristles on the flexor surfaces of the tibiae and tarsi and a few blunt hairs on the anterior surfaces of the fore coxae.

Sordid brownish red, the smaller specimens somewhat more yellowish red. Gaster dark brown, except a large spot at the base of the first segment and the anal region, which are reddish yellow. A large spot on the pronotum, one on the mesonotum, much of the posterodorsal portion of the head, the distal halves of the antennal funiculi and in many specimens also the coxae and femora, dark brown or blackish. These dark markings are present in the largest as well as in the smallest workers.

HOST (TEMPORARY). Probably *F. fusca*.

TYPE LOCALITY.— Michigan: Isle Royale (H. A. Gleason).

In coloration, this ant resembles very closely small specimens of the European *F. rufa pratensis*, and can be distinguished from all the preceding forms of the *microgyna* group by the extensive infuscation of the upper surface of the head. Mr. Gleason describes the nests on Isle Royale as "one of the most conspicuous features of the drier tamarack swamps. They are rounded-conical in shape, 3–6 dcm. high or even larger, with a diameter at the base about equalling the height. They are composed within of Sphagnum, but as would be expected with such material, without any definite system of galleries. The outer surface is thickly covered with leaves of Cassandra, probably to prevent loss of moisture by evaporation from the interior. They are frequently placed near or under a bush of the Cassandra, but the same covering is used if no Cassandra is near."

61. F. ADAMSI var. ALPINA Wheeler.

F. adamsi var. *alpina* Wheeler, Journ. N. Y. ent. soc., 1909, **17**, p. 84, ☿; Rept. Mich. geol. survey for 1908, 1909, p. 327, ☿.

Worker. Length 3.5–5 mm.

Differing from the typical *adamsi* in having the border of the petiole more attenuated and more produced upward in the middle, in the black markings on the head, pro- and mesonotum being more restricted and in having the frontal area smoother and more shining.

Type locality.— Colorado: Pikes Peak, 10,500–11,000 ft., (Wheeler).

Idaho: Troy (W. M. Mann).

Nova Scotia: Boisdale, Cape Breton I. (Amer. Mus. Nat. Hist. Coll.).

The red portions of the specimens from Idaho are paler than in those from Colorado and Cape Breton I. and the yellow spot at the base of the gaster is conspicuous. The true status of this variety, however, can be determined only by the study of more material than I have been able to secure.

62. F. NEPTICULA Wheeler.

F. nepticula Wheeler, Bull. Amer. mus. nat. hist., 1905, **21**, p. 270, ☿ ♀ ♂; Ibid., 1906, **22**, p. 64.

Worker. Length 3.5–6 mm.

Mandibles 8-toothed. Head, excluding the mandibles, a little longer than broad, but little narrower in front than behind, with straight sides and posterior border. Clypeus strongly carinate, its anterior border angularly produced in the middle. Frontal area triangular, as long as broad. Antennae rather stout, first to fourth funicular joints longer and more slender than the penultimate joints. Thorax in profile with very convex pro- and mesonotum and very deep mesoëpinotal constriction, which is broad at the bottom. Epinotum rounded, without distinct base and declivity, or, at any rate, without an angle between the base and declivity. Petiole large, as high as the epinotum, convex in front, more flattened behind, border rather sharp; seen from behind it is transverse in the middle and obliquely truncated on each side, the lateral borders being straight and converging below.

Head, thorax, and petiole subopaque, very finely shagreened; mandibles, clypeus, and frontal portion of head, and especially the frontal area, more shining. Mandibles densely striated and coarsely punctate. Legs and gaster shining, the latter delicately and transversely shagreened, with the same peculiar luster as in *F. truncicola obscuriventris.*

Hairs golden yellow, short, obtuse, suberect and very sparse on the upper and lower surfaces of the head, thoracic dorsum, and gaster. Legs without erect hairs on the extensor surfaces; antennal scapes occasionally with a few short hairs on their anterior surfaces. Eyes hairless. Pubescence whitish, very short and sparse, visible on the antennae, pleurae, and gaster, but not concealing the shining surface of the gaster.

Mandibular teeth and gaster black; remainder of body and appendages deep red; antennal funiculi, legs, especially the tibiae, mandibles, and anterolateral corners of the head, darker and more brownish. Ocellar region and mesonotum slightly infuscated even in large workers, but there is no increased tendency to infuscation in the smaller workers.

FEMALE. Length 4–5 mm.

Mandibles and clypeus like those of the worker, except that the latter is more convex and less prominently keeled. Head slender, without the mandibles distinctly longer than broad, with long, anteriorly converging cheeks. Thorax distinctly narrower than the head. Petiole similar to that of worker but with sharper superior border, often slightly notched in the middle. Gaster small. Legs slender. Wings somewhat longer than the body (5.3 mm.).

Body smooth and shining, very finely shagreened, posterior portion of head and mesonotum more opaque; gaster very glabrous, being much more delicately shagreened than in the worker.

Hairs golden yellow, suberect, slender, pointed, much longer than in the worker and more abundant, especially on the upper surface of the head and thorax. Legs with rather long, scattered, subappressed hairs. There are a few conspicuous erect hairs on the anterior surface of the antennal scapes, on the gula and border of the petiole. On the gaster the long hairs are sparse and arranged in three regular rows on the first and second, in two rows on the succeeding segments.

Mandibular teeth and gaster black, remainder of body dull yellowish red. Antennae, legs, posterior portion of head, mesonotum, scutellum, and metanotum decidedly darker. The anteromedian and parapsidal blotches are faintly indicated on the mesonotum. Wings rather opaque, grayish hyaline, with fuscous veins and black stigma.

MALE. Length 6.5–7 mm.

Mandibles pointed, edentulous. Head short, broadest through the eyes, which are large. Posterior corners broadly rounded; cheeks short, flattened, converging in front. Clypeus carinate in front, depressed behind. Thorax just in front of the wings hardly broader than the head through the eyes. Base of epinotum with a median longitudinal impression, metanotum concave. Petiole very thick and blunt above, anterior and posterior surfaces both convex, border with a faint median notch.

Head, thorax, legs, and antennae subopaque, finely shagreened; mandibles, clypeus, frontal area, vertex, and scutellum shining as are also the petiole and especially the gaster.

Hairs and pubescence grayish, the former short and erect on the clypeus, thorax, gaster, and legs; the latter sparse and indistinct except on the antennae and legs. Eyes almost imperceptibly hairy.

Black; mouthparts, legs, and genitalia fuscous. Wings like those of the female but of a slightly darker tint.

HOST (TEMPORARY). Probably *F. neogagates.*

TYPE LOCALITY.— Connecticut: Colebrook, 1,400 ft. (Wheeler).

Massachusetts: Stony Brook Reservation, Chestnut Hill, near Boston (Wheeler).

Illinois: Black Hawk Springs, near Rockford (Wheeler).

The female *nepticula* resembles the female *nevadensis,* but differs in having much fewer erect hairs on the antennal scapes and body and, owing to the nearly complete absence of grayish pubescence, a more shining head and thorax. Moreover, the head, thorax, and appendages are decidedly darker and less red than in *nevadensis.* The worker *nepticula* may be readily confounded with that of *F. truncicola obscuriventris* owing to both forms having the same color and the same luster of the gaster, but *nepticula* is of average smaller size, has much sparser, coarser, and more obtuse hairs, the border of the clypeus is more projecting, and the epinotum is much lower and rounder.

F. nepticula is, in my experience, a rare ant. It nests in open woods under stones, the edges of which it banks with vegetable detritus. The colonies are rather small. The males and diminutive females make their appearance early in July.

63. F. DIFFICILIS Emery.

F. pallidefulva Mayr (*nec* Latreille), Verh. Zool. bot. ver. Wien, 1866, **16,** p. 889, ♀.

F. rufa subsp. *difficilis* Emery, Zool. jahrb. Syst., 1893, **7**, p. 651, pl. 22, figs. 9, 14, ☿ ♀ ♂.

F. difficilis Wheeler, Bull. Amer. mus. nat. hist., 1904, **20**, p. 348; Ibid., 1906, **22**, p. 63.

WORKER. Length: 3.5–5.5 mm.

Head, excluding the mandibles, slightly longer than broad, slightly broader behind than in front, with feebly convex posterior and lateral borders. Clypeus carinate its entire length, with the anterior border angularly projecting. Mandibles 8-toothed. Frontal area triangular, as long as broad. Frontal carinae strongly diverging. Antennae slender, basal funicular joints longer and more slender than the apical joints. Maxillary palpi rather short. Pro- and mesonotum not very convex, mesoëpinotal constriction rather shallow, base of epinotum convex, declivity flat, very sloping, and forming a rounded obtuse angle with the base. Petiole narrow, thick, with more convex anterior, and less convex posterior surface and blunt border, which when seen from behind is broadly rounded and not produced upward in the middle. Gaster of the usual shape; legs rather slender.

Opaque; mandibles, clypeus, and front of head slightly shining; frontal area subopaque. Mandibles very finely and densely striated.

Erect hairs very sparse, short, blunt, clavate, present on the front, clypeus, ocellar region, gula, upper surface of the pro- meso- and epinotum, and gaster. Antennal scapes and legs without oblique or suberect hairs. Pubescence grayish, sparse and fine on the head, thorax and appendages, denser on the gaster and concealing the surface so that this region appears gray.

Head, thorax, and petiole bright red or orange red; gaster dark brown with the base of the first segment and the anal region red or yellow. In small specimens the posterodorsal portions of the head and the dorsal portions of the thorax and petiole are more or less infuscated. Even in the largest workers the ocellar triangle is fuscous.

FEMALE. Length 4–5.5 mm.

Head very much like that of the worker. Whole surface of body smoother and a little more shining. Hairs much longer and more abundant, present also on the posterolateral corners of the head. Pubescence yellow, very fine but distinctly visible on the upper surface of the head and thorax, and thick enough on the gaster to obscure the shining surface.

Reddish yellow throughout, only the eyes, mandibular teeth and wing insertions black. Wings grayish hyaline, more infuscated towards their bases. Veins and stigma pale brown.

MALE. Length 5.5–6 mm.

(After Emery). Mandibles strongly dentate. Head short, broadest through the eyes which are large, behind the eyes broadly convex. Black; antennal funiculi usually brown, mandibles, legs, and genitalia paler yellow, femora often daker, coxae brown. Wings pale grayish, with dark veins.

HOST (TEMPORARY). Probably *F. schaufussi* or one of its varieties.

TYPE LOCALITY.— Virginia: (Th. Pergande).

North Carolina: Black Mts., Swannanoa Valley (W. Beutenmüller).

New Jersey: Lakehurst, Halifax (Wheeler); Brown's Mills Junction (W. T. Davis).

New York: Bronxville (Wheeler).

This ant, originally described as a form of *rufa*, occurs sporadically in open mountainous woods from New York state to North Carolina and probably somewhat further south along the Alleghanies. It nests under stones, which it banks with vegetable detritus. The colonies are often moderately large.

64. F. DIFFICILIS var. CONSOCIANS Wheeler.

F. difficilis var. *consocians* Wheeler, Bull. Amer. mus. nat. hist., 1904, **20**, p. 371, ☿ ♀ ♂; Ibid., 1906, **22**, p. 50.

WORKER. Length 3.5–5.5 mm.

Closely resembling the typical form, but the erect hairs more abundant and slightly longer, especially on the front, gula, and thorax. There are also numerous hairs on the posterolateral corners of the head, which are nearly always lacking in the typical *difficilis*. The petiolar border is somewhat sharper and the frontal area is smoother and more shining.

FEMALE. Length 4–5.5 mm.

Differing from the female of the typical *consocians* in having the pubescence and pilosity more abundant. The former is rather dense so that the whole body except the anterior portion of the head appears much less shining. The tibiae have long, scattered, oblique or suberect hairs which are lacking in the typical *difficilis*. Wings grayish hyaline, darker at the base.

MALE. Length 5.5–6.5 mm.

Mandibles broad, usually edentate, but occasionally with minute teeth at the base. Clypeus sharply carinate. Petiole thick, transverse, its anterior surface angularly convex, its posterior surface more flattened, its border obtuse, seen from behind broadly rounded and entire.

Body subopaque, head and gaster somewhat more shining. Mandibles coarsely punctate. Frontal area smooth and shining.

Hairs yellowish, erect, rather abundant on the head, mesonotum and petiole, sparse on the pleurae and upper surface of the gaster. Tibiae with a few small oblique hairs. Eyes hairless. Pubescence rather long and conspicuous on the thorax and gaster, shorter and sparser on the head, dense and very fine on the legs.

Head and thorax black; mandibles, antennae, petiole, and gaster dark brown; legs and genitalia light yellow; fore femora sometimes slightly infuscated. Wings grayish hyaline, distinctly infuscated towards their bases; veins and stigma brown.

HOST (TEMPORARY). *F. schaufussi* var. *incerta.*

TYPE LOCALITY.— Connecticut: Colebrook, (Wheeler).

Massachusetts: Woods Hole, Ellisville, Forest Hills, Blue Hills (Wheeler).

The habits of this variety are very similar to those of the typical form. Its young colonies are not infrequently found mixed with *F. incerta* and, as I have shown in former papers, the recently impregnated queens establish their formicaries with the aid of this species. The characters which separate *consocians* from the typical form are very slight and refer mostly to the pilosity. I believe, however, that they will prove to be valid. The variety is evidently a more boreal form of the species.

65. F. MORSEI Wheeler.

F. morsei Wheeler, Psyche, 1906, **13**, p. 39, fig. ☿.

WORKER. Length 3.5–5.5 mm.

Mandibles 8-toothed. Palpi rather long. Head, excluding the mandibles, distinctly longer than broad; cheeks long, slightly flattened converging in front; posterior border and angles convex and rounded. Clypeus convex, carinate, with rounded anterior border. Antennae slender; four basal joints of funiculi longer and more slender than the penultimate joints. Thorax in profile with deep mesoëpinotal constriction, convex pro- and mesonotum and the epinotum with distinct base and declivity, the former convex and rounded, the latter flattened and sloping. Petiole narrow, its anterior and posterior surfaces alike convex in profile, its border rather blunt; seen from behind the border is broadly rounded, in some specimens faintly excised on the middle, but not produced upward. Gaster large. Legs rather long.

Mandibles subopaque, coarsely striatopunctate. Anterior portion of head, clypeus, frontal area, lower surface of thorax, and gaster smooth and shining; remainder of body subopaque, very finely shagreened; upper surface of gaster with a slightly oily luster.

Hairs white, short, obtuse, suberect, and very sparse on the upper surface of the head, thorax, and gaster; nearly always absent on the gula and petiolar border. Femora and tibiae naked, the latter with a row of tapering hairs on their flexor surfaces. Pubescence white, extremely short and sparse, so that it is almost invisible, except on the upper surface of the gaster.

Reddish yellow; borders of mandibles black; anterior border of clypeus, vertex, upper surface of pro- and mesonotum, femora, tibiae, apical antennal joints, and gaster more or less infuscated; anal region yellow. In many specimens the upper surface of the head is more reddish than the remainder of the body but there is little difference in coloration between the smallest and largest workers.

TYPE LOCALITY. — Massachusetts: South Natick (A. P. Morse).

The position of this species is problematic, as the female is unknown, but the characters of the worker certainly ally it to the preceding forms of the *microgyna* group. It differs from all of these species in its peculiar color and sculpture, the greater convexity of the posterior portion of the head and the shape of the petiole. It must be a rare species as I have been unable to find it again, even with Mr. Morse's assistance, in the type locality.

Exsecta Group.

66. F. EXSECTOIDES EXSECTOIDES Forel.

F. integra Mayr, Verh. Zool. bot. ver. Wien, 1862, **12**, p. 70; Ibid., 1886, **36**, p. 425, ♀ ♂ (*nec* Nylander).

F. exsectoides (Forel) McCook, Trans. Amer. ent. soc., 1877, **6**, p. 252–296, figs. 2–5, pls. 2–6; Proc. Acad. nat. sci. Phila., 1877, p. 135; Amer. natur., 1878, **12**, p. 431–345, 8 figs.; Proc. Acad. nat. sci. Phila., 1879, p. 154–156.

F. exsectoides Forel, Ann. Soc. ent. Belg., 1886, **30**, C. R. p. xxxviii, ☿ ♀; Dalla Torre, Catalog. Hymen., 1893, **7**, p. 195; Emery, Zool. jahrb. Syst. 1893, **7**, p. 653, pl. 22, fig. 6; Wheeler, Bull. Amer. mus. nat. hist., 1906, **22**, p. 71, 403–413, pls. 43–48.

WORKER. Length 4.5–7.5 mm.

Mandibles with the apical border 8-toothed, the basal border with two or three minute and often very indistinct or obsolete denticles. Head, excluding the mandibles as broad as long, a little narrower in front than behind, with deeply excised posterior and nearly straight lateral borders. Front convex, vertex flattened. Clypeus strongly carinate, with anterior border entire and angularly projecting in the middle. Frontal area distinct, frontal carinae moderately diverging. Antennae slender, scapes not thickened towards their tips, funiculi with the four basal joints somewhat longer and more slender than the penultimate joints. Maxillary palpi moderately long, 6-jointed. Pro- and mesonotum distinctly flattened above, mesoëpinotal constriction rather shallow; epinotum with subequal base and declivity, each straight in profile and meeting at a rounded obtuse angle. Petiole high and broad, compressed anteroposteriorly, with flattened anterior and posterior surfaces, and sharp, cultrate border, which, seen from behind, is entire and usually broadly rounded, or slightly produced upward in the middle. Gaster of the usual shape, legs rather long.

Body delicately shagreened; head, thorax, and petiole subopaque or slightly shining; mandibles and frontal area more shining, the former striatopunctate, the latter rather smooth; gaster shining but more distinctly shagreened than the head and thorax.

Hairs and pubescence yellowish, very sparse. There are a few hairs on the clypeus, mandibles, and sometimes on the front, and blunt scattered hairs on the gaster. Eyes hairless. Pubescence short and very dilute, most distinct on the gaster. Legs without erect hairs, invested with very fine, dilute pubescence.

Deep red; gaster black, anal region reddish; mandibles, legs, vertex, funiculi and dorsal portions of thorax sometimes brownish or dark red.

Female. Length: 9–11 mm.

Head resembling that of the worker, but more flattened at the vertex, scarcely broader than the thorax. Petiole broader than that of the worker but similar in shape.

Head and thorax somewhat more shining than in the worker; mandibles superficially striated and coarsely punctate.

Hairs tawny, long, coarse, flexuous, and sparse, confined to the posterior portion of the pronotum and the gaster. There are also two small tufts of these hairs on the mesonotum, and several on the scutellum and clypeus. The pubescence is even more feebly developed than in the worker, being almost absent on the gaster.

Color like that of the worker, except that the gaster has a more reddish tinge and a yellow spot at the base of the first segment. Wings uniformly brown, with brown veins and stigma.

Male. Length 7.5–10 mm.

Mandibles with broad blades, distinctly 4-toothed. Eyes large. Head broad behind, with straight posterior border and large posterior corners, narrow in front, with short, straight cheeks. Clypeus sharply carinate, with broadly rounded anterior border. Maxillary palpi 5-jointed. Thorax robust. Petiole a little higher than long, thick, with flattened anterior and posterior surfaces, and very blunt, entire superior border.

Head, mandibles, thorax, and petiole subopaque; gaster shining. Mandibles coarsely striato-punctate.

Hairs few and scattered, confined to the upper surface of the head and thorax. Pubescence long, grayish, conspicuous and rather dense on the head, thorax, and gaster.

Black; mandibles, and antennae brown, genitalia brownish yellow; legs, including the coxae, light yellow. Wings brown as in the female, with brown veins and stigma.

HOST (TEMPORARY). *F. fusca* var. *subsericea.*

TYPE LOCALITY.— New Hampshire (Forel).

New Hampshire: Canobie Lake (G. B. King); Franconia (Mrs. A. T. Slosson); Raymond (W. Reiff).

Georgia: Rabun Bald Mountain (W. T. Davis).

North Carolina: Black Mountain (Forel).

Maryland: Baltimore (E. A. Andrews); Prince George County (W. T. Davis).

New Jersey: Newfoundland (W. T. Davis and Wheeler); Palisades; Alpine (W. Beutenmüller); Westfield, Scotch Plains, Halifax, Paterson (Wheeler); Tenafly (G. v. Krockow).

Pennsylvania: Hollidaysburg, Warrior's Mark, etc., (H. C. McCook); Lehigh Water Gap, Beatty (P. J. Schmitt).

New York: Staten Island (W. T. Davis); Ramapo Mts., Bronxville (Wheeler); West Farms (J. Angus); Garrison-on-Hudson (T. D. A. Cockerell).

Connecticut: Branford, North Haven, New Haven (H. L. Viereck); New Hartford, Stafford (W. E. Britton); Cromwell, Hartford (Forel); Colebrook (Wheeler).

Massachusetts: Sherborn, Wellesley (A. P. Morse); Essex County, Mt. Tom (G. B. King); Lowell, Tyngsboro (F. Blanchard); Lake Pleasant (Carey); Warwick (Miss Edwards); Woods Hole, Forest Hills, Blue Hills (Wheeler); Worcester (Forel).

Maine: Ogunquit (H. S. Pratt); South Harpswell (Wheeler).

Illinois: (M. C. Tanquary).

Wisconsin: Prairie du Chien (H. Muckermann).

Ontario: Toronto (R. J. Crew).

Nova Scotia: Round Hill (Centr. Exp. Farms Coll.).

This is the well-known "mound-building ant of the Alleghenies," the habits of which were described many years ago by Rev. H. C. McCook, who studied its huge colonies (one of them comprising some 1,600 nests!) in the mountains of Pennsylvania. The nests are large conical mounds, often 2.5 ft. high and 9.5 ft. in convex diameter, consisting very largely of earth, and erected in clearings in the woods. I have shown that the females establish their colonies by temporary parasitism in small colonies of *F. fusca* var. *subsericea.* Old colonies are frequently extinguished or compelled to move to new quarters by the growth of a carpet of moss (*Polytrichum commune*) over the surface of the nest. *F. exsectoides* is a very fierce ant and furiously attacks any intruder on its preserves. It kills other ants by decapitating them, a habit which seems to be peculiar to the members of the *exsecta* group.

67. F. EXSECTOIDES EXSECTOIDES var. DAVISI, var. nov.

WORKER. Length 4.5–7.5 mm.
Differing from the worker of the typical form only in having the posterior portion of the head and the dorsal portion of the pro- and mesonotum distinctly infuscated, at least in many of the workers of all sizes in the colony.
FEMALE (DEÄLATED). Length 9–11 mm.
Gaster red like the thorax and head and transversely banded with black, owing to the anterior and posterior border of each segment being of this color.

Described from a number of workers and deälated females taken at Newfoundland, N. J., by Mr. Wm. T. Davis. I have also taken this same form at Natick, Mass. Its validity as a variety will have to be tested by further study of the species. Possibly the color of the gaster in the queens is due to old age. It is, however, constant in twenty-one specimens taken from twelve different nests in New Jersey and fully thirty females from as many nests in Massachusetts.

68. F. EXSECTOIDES EXSECTOIDES var. HESPERIA, var. nov.

WORKER. Length 4.5–6 mm.
Differing from the typical form in the shape of the petiole, which is much narrower, lower and thicker, with the anterior surface convex, the posterior flattened, and the border, though sharp, not being blade-like, or cultrate. Seen from behind it is truncated and like the petiole of *F. dakotensis* in outline. The posterior corners of head, ocellar triangle, and a spot on the pro- and mesonotum fuscous. The red color of the body is a little more brownish and the legs darker than in the typical form. Frontal area rather opaque.

Described from twenty-eight workers which I took from a single colony nesting under a cluster of stones in Cheyenne Canyon, near Colorado Springs, Colo.

69. F. EXSECTOIDES OPACIVENTRIS Emery.

F. exsectoides var. *opaciventris* Emery, Zool. jahrb. Syst., 1893, **7**, p. 653, ☿; Wheeler, Bull. Amer. mus. nat. hist., 1906, **22**, p. 405.

WORKER. Length 4.5–6 mm.
Differing from the typical *exsectoides* in having the antennal scapes distinctly thickened at their tips and in the greater abundance of the

hairs and pubescence. There are prominent golden yellow hairs on the clypeus, front, and vertex and on the dorsum of the pro- and mesonotum, and the hairs on the gaster, which are also golden or fulvous, are coarser. The pubescence is grayish and denser on all parts of the body but especially on the gaster. This region is also more coarsely shagreened and therefore subopaque or opaque and not shining. The funiculi and the tips of the scapes are fuscous, in other respects the color is like that of the typical form.

Male. Length 7.5 mm.

Differing from the male of the typical *exsectoides* in having the thorax and gaster invested with longer pubescence and the mesonotum and scutellum covered with more abundant subappressed hairs. Eyes hairy. The single rather immature specimen examined has bidentate mandibles. The petiole is broadly excised in the middle and its margin has the form of a pointed, compressed tooth on each side of the excision.

Type locality.— Colorado: Breckenridge (Emery).

Colorado: Boulder (P. J. Schmitt and Wheeler); Florissant (Wheeler).

This is, in my opinion, a sharply marked subspecies and not a mere variety. It is readily distinguished from the typical form also in its habits. Its nests resemble those of the eastern form in size and shape, but are made of earth and pebbles instead of earth and vegetable detritus. At least this was the case with several nests which I saw at Florissant. When these nests were situated near the railroad track they were covered also with locomotive cinders which the ants had carefully collected. All the nests examined were in open country, not in woods.

70. F. ULKEI Emery.

F. ulkei Emery, Zool. jahrb. Syst., 1893, **7**, p. 653, pl. 22, fig. 7, ☿; Wheeler, Ants, 1910, p. 446, 570.

Worker. Length 3.5–6 mm.

Mandibles with 8 teeth on the apical and a few indistinct denticles on the basal margin. Head, excluding the mandibles, distinctly longer than broad, very slightly narrower in front than behind, with broadly and deeply excised posterior border and rather straight sides. Front convex, vertex less flattened than in *exsectoides*. Clypeus carinate, but less acutely than in *exsectoides*, its anterior margin, angularly produced. Frontal area triangular, a little broader than long; frontal carinae at first diverging posteriorly but at their posterior ends more nearly parallel. Maxillary palpi rather long, 6-jointed. Antennal scapes distinctly incrassated at their tips; joints 2–4 of the funiculi a little more slender, but scarcely longer than the penultimate joints. Pro- and mesonotum distinctly depressed or flattened above; mesoëpinotal impression shallow and broad at the bottom; epinotum with the base a little shorter than the declivity, the former slightly convex in profile and passing through a rounded angle into the sloping declivity. Petiole high and rather broad, much compressed anteroposteriorly, the anterior surface somewhat convex, the posterior flat, the upper border cultrate and sharp; seen from behind the sides are straight and diverge upward and the superior border is horizontal and entire.

Surface of body very finely shagreened, head and gaster shining, thorax subopaque; mandibles finely striatopunctate; frontal area subopaque; surface of gaster finely and sparsely punctate and transversely shagreened, with the sheen of the gaster of *exsectoides, obscuriventris,* and *nepticula,* but slightly less shining.

Hairs golden yellow, sparse and coarse, present on the clypeus, front, vertex, pro- and mesonotum, fore coxae, and gaster; slightly shorter on the gaster than in *exsectoides,* long on the venter. Pubescence very fine and sparse, most clearly visible on the gaster and legs.

Light or dark ferruginous red, legs a little darker, and more brownish; gaster, posterior half of head, a large spot on the pronotum, and a small one on the mesonotum, black.

FEMALE. Length 7.5–9 mm.

Smaller than the female of *exsectoides.* Resembling the worker in color, but the tips of the scapes, the pleurae, bases of fore coxae and three large spots on the thorax are dark brown, and the surface of the body, especially of the head and thorax, is more glabrous and shining. The clypeus is ecarinate, with the anterior border depressed, the mandibles rather superficially striatopunctate.

The pilosity of the head is like that of the worker, but the hairs on the remainder of the body are very different. The pronotum, mesonotum, scutellum, upper pleurae, and gaster are covered with sparse, very long and coarse, appressed, tawny hairs. The border of the petiole also bears a number of these peculiar hairs. Wings uniformly tinged with brown, with pale brown veins and stigma.

MALE. Length 7–8 mm.

Head like that of the male *exsectoides,* with straight posterior border, but with somewhat more broadly elliptical eyes. Mandibles edentate, narrow, and pointed. Thorax and gaster, robust, the latter flattened and rather broad. Petiole thick and low, with blunt, entire, transverse border.

Whole body more shining and more finely shagreened than in *exsectoides.*

Pubescence grayish, much shorter and sparser, especially on the head and thorax. Hairs of the same color, but longer and more abundant than in *exsectoides,* though restricted to the upper surface of the thorax, praesternum, and tip of gaster. Eyes hairless.

Head, including the mandibles and antennae, thorax, petiole, and gaster of a deeper black than in *exsectoides;* legs and genital appendages of a more reddish yellow, with the bases of the femora on the flexor surface and the tips of genital valves blackish. Wings slightly paler than in *exsectoides.*

HOST (TEMPORARY). Probably *F. fusca.*

TYPE LOCALITY.— South Dakota: Hill City (T. Ulke).

Illinois: Chicago (M. C. Tanquary).

Nova Scotia: Bedford, Port Maitland (W. Reiff); Middleton, Round Hill (Centr. Exper. Farms Coll.); Delhaven (Cornell Univ. Coll.); Ship Harbor (S. Henshaw).

New Brunswick: Fredericton (J. D. Tothill).

The female and male are described from Nova Scotia and New Brunswick specimens respectively. This species is evidently peculiar to the Canadian fauna and so rare in the transition zone that I have never had the good fortune to find one of its colonies. Mr. J. D. Tothill, who has been studying its habits in New Brunswick, has kindly given me photographs and a description of its nests. These are flattened mounds, a foot or somewhat more in diameter, made of earth and considerable vegetable detritus, and therefore seem to be much more like the nests of *exsecta* than those of *exsectoides.* In the coloration of the worker, the shape of its head, the small size of the female and the sculpture of the male, *ulkei* also approaches the European species, but its strongest morphological affinities are nevertheless with *exsectoides.*

71. F. ULKEI var. HEBESCENS, var. nov.

WORKER. Length 4.5–6 mm.

Differing from the typical form in sculpture and coloration. The shagreening of the gaster is much sharper so that this region is subopaque or only slightly shining. The anterior half of the head and the thorax, petiole, and legs are more brownish red than in the typical form, while the gaster and posterior half of the head are brown instead of black, and the spots on the thorax are paler.

TYPE LOCALITY.— Indiana: Bass Lake, Stark County (W. S. Blatchley).

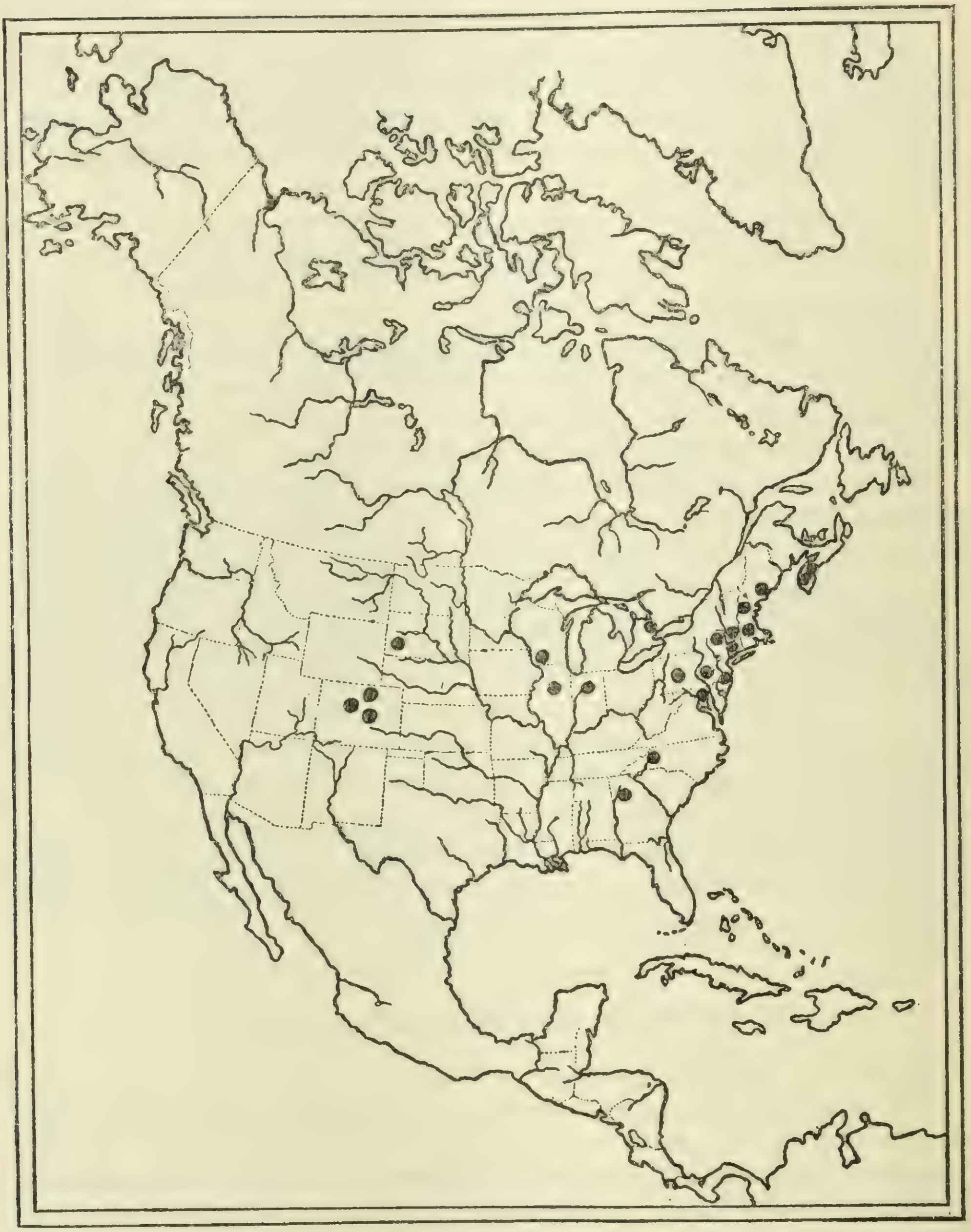

FIG. 5.— Distribution of the Nearctic forms of the *Formica exsecta* group.

Indiana: Tippecanoe Lake (W. S. Blatchley).

Nova Scotia: Digby (J. Russell).

Described from a large number of specimens, all agreeing in the characters noted above.

72. F. EXSECTA EXSECTA Nylander.

F. exsecta Nylander, Acta Soc. Fennica, 1846, **2**, p. 909, pl. 18, fig. 20, ☿ ♀ ♂; Ann. sci. nat. Zool., 1856, ser. 4, **5**, p. 63, pl. 3, fig. 7, ☿ ♀ ♂; Förster, Hymen. stud., 1850, **1**, p. 23, ☿ ♀ ♂; Mayr, Verh. Zool. bot. ver. Wien, 1855, **5**, p. 340, ☿ ♀ ♂; Europ. Formicid., 1861, p. 46–48; Forel, Denks. Schweiz. gesell. Naturw., 1874, **26**, p. 51, ☿ ♀ ♂; Ern. André, Spec. Hymén. Europ., 1882, **2**, pt. 14, p. 178, 185, 188; Dalla Torre, Catalog. Hymen., 1893, **7**, p. 195; Ruzsky, Formicar. Imper. Ross., 1905, p. 353, figs. 67, 68; Emery, Deutsch. ent. zeitschr., 1909, p. 189, fig. 5; Ibid., 1912, p. 672.

WORKER. Length 5–7.5 mm.

Mandibles 8-toothed, with additional indistinct denticles on the basal margin. Head longer than broad, narrowed somewhat at the posterior corners so that it is here no broader than at the anterior border; posterior border deeply excised. Front convex; clypeus sharply carinate, without a distinct, transverse depression behind its anterior border. Maxillary palpi long, 6-jointed. Pro- and mesonotum not very convex; mesoëpinotal constriction shallow, broad at the bottom. Spiracles of metanotum prominent. Epinotum rounded, without distinct base and declivity. Petiole narrow, strongly compressed anteroposteriorly, its superior margin thin, sharp, and very deeply excised.

Opaque or slightly shining. Mandibles finely striated and coarsely punctate.

Hairs very sparse, distinct on the gaster, where they are short and obtuse. Pubescence yellow, moderately abundant, especially on the head, prothorax, and gaster, longest on the cheeks. Eyes hairless.

Red or sordid yellowish red; clypeus and appendages darker, posterodorsal portion of head, a large spot on the pronotum and often also a small one on the mesonotum, brown; gaster blackish brown, base of first segment red or yellow.

FEMALE. Length 8–9.5 mm.

Resembling the worker. Anterior border of clypeus straight in the middle. Thorax with flattened mesonotum and scutellum. Petiole much broader, flatter, and more deeply excised than in the worker.

Sculpture much as in the worker. Hairs more abundant especially on the mesonotum, posterior border of pronotum, and front of head. Pubescence longer but sparse, conspicuous both on the body and appendages.

Ferruginous red; top of head, posterior border of pronotum, the mesonotum, scutellum, metanotum, and gaster dark reddish brown. Wings brownish hyaline, with pale brown veins and stigma.

MALE. Length 6–9 mm.

Head rather small, with broadly excised posterior margin. Eyes large. Mandibles pointed, edentate, and rather narrow. Maxillary palpi as in the worker and female. Petiole low, somewhat compressed, with blunt, broadly excised border.

Body rather shining, especially the gaster.

Hairs shorter than in the female, most abundant and distinct on the head and thoracic dorsum. Eyes hairy. Pubescence short, most distinct on the upper surface of the gaster.

Black; legs and genitalia brown, femora somewhat darker. Wings colored as in the female.

HOST (TEMPORARY). *F. fusca.*

North and Middle Europe; Alps, Caucasus, Siberia, Altai Mountains.

Lives in woods and builds flattened mound nests covered with finer plant débris than those of *rufa* and *pratensis*. Single colonies often occupy more than 100 nests which are connected with one another by runways and may cover a considerable area.

73. F. EXSECTA EXSECTA var. RUBENS Forel.

F. exsecta var. *rubens* Forel, Denks. Schweiz. gesell. naturw., 1874, **26**, p. 51, ☿; Ern. André, Spec. Hymén. Europe, 1882, **2**, pt. 14, p. 179; Dalla Torre, Catalog. Hymen., 1893, **7**, p. 195; Ruzsky, Formicar. Imper. Ross., 1905, p. 358; Emery, Deutsch. ent. zeitschr., 1909, p. 191, ☿.

WORKER. Differs from the typical *exsecta* only in color, the body being pale red, with a small spot on the vertex and the gaster, except its base, brown.

This form is known only from the Swiss Jura and Southern Russia. I am inclined to include under it also a number of workers which I took at Zermatt, Switzerland. In these the spot on the head is not smaller than in the typical form but much paler.

74. F. EXSECTA EXSECTA var. ETRUSCA Emery.

F. exsecta var. *etrusca* Emery, Deutsch. ent. zeitschr., 1909, p. 191, ☿.

WORKER. Length 5–6.5 mm.

Maxillary palpi a little shorter than in the typical *exsecta*. Dark red, the brown color on the head more extensive. Legs, or at least the tibiae, brown. Petiole remarkably broad, its superior border rounded, entire or only slightly excised.

Italy: Pracchia and Abetone in the Apennines (C. Emery).

75. F. EXSECTA PRESSILABRIS Nylander.

F. pressilabris Nylander, Acta. Soc. Femnica, 1846, **2**, p. 911, pl. 18, fig. 21, ☿ ♀ ♂; Meinert, Naturv. abh. Dansk. vid. selsk. (5) **5**, 1860, p. 45, ☿ ♀ ♂; Dalla Torre, Catalog. Hymen., 1893, **7**, p. 205, (*in part*).

F. exsecta subsp. *pressilabris* Ruzsky, Formicar. Imper. Ross., 1905, p. 363, figs. 69, 70; Emery, Deutsch. ent. zeitschr., 1909, p. 191, fig. 5; Ibid., 1912, p. 672.

WORKER. Length 3.8–6.5 mm.

Maxillary palpi very short, 5-jointed, or 6-jointed, according as the fourth joint is more or less distinctly separated into two small joints. Clypeus with a transverse impression just behind and parallel with its somewhat reflected anterior border. Front convex anteriorly, depressed behind.

Sculpture very fine and superficial. Gaster, especially at the base, lustrous.

Pubescence very short and sparse.

Red portions of body darker than in the typical *exsecta,* the dark spots more extensive, antennae and legs often brown.

FEMALE. Length 6–7.5 mm.

Much smaller than the female of the true *exsecta* and very shining. Palpi as in the worker. Pubescence extremely short and sparse. Dark brown, anterior portion of head, ventral and lateral portions of thorax, mesonotum in part, petiole below, and tip of gaster, often also the femora and the scapes, paler or darker red.

MALE. Length 5–7.5 mm.

Smaller than the male of the typical *exsecta;* the posterior margin of the head more feebly excised; maxillary palpi as in the worker; eyes hairless.

HOST (TEMPORARY). *F. fusca.*

Northern Europe, Caucasus, Siberia, Turkestan, Ural Mountains.

76. F. EXSECTA PRESSILABRIS var. FORELI Emery.

F. pressilabris Mayr, Verh. Zool. bot. ver. Wien, **5**, 1855, p. 339, ☿ ♀ (*nec* ♀); Europ. Formicid, 1861, p. 46; Dalla Torre, Catalog. Hymen., 1893, **7**, p. 205 (*in part*).

F. exsecta st. *pressilabris* Forel, Denks. Schweiz. gesell. naturw., 1874, **26**, p. 55, ☿ ♀ ♂.

F. exsecta pressilabris var. *foreli* Emery, Deutsch. ent. zeitschr., 1909, p. 192, ☿ ♀ ♂.

Worker.

Differing from *pressilabris* in its less superficial sculpture and somewhat longer pubescence. The upper surface of the gaster is quite opaque at the base.

Female.

Somewhat more shining than the female of the typical *exsecta;* pubescence much longer and denser.

Male.

Indistinguishable from the male of *pressilabris.*

Distributed through Switzerland and probably also through the mountain regions of Central Europe. The nest mounds are small and more earth is used in their construction than in the nests of the typical *exsecta.* They are most frequently found in meadows, especially along the borders of hedges and woods. A single colony often inhabits several nests.

77. F. EXSECTA PRESSILABRIS var. EXSECTO-PRESSILABRIS Forel.

F. exsecta var. *exsecto-pressilabris* Forel, Denks. Schweiz. gesell. naturw., 1874, **26**, p. 52, 55, 57, ☿ ♀ ♂; Dalla Torre, Catalog. Hymen., 1893, **7**, p. 205; Emery, Deutsch. ent. zeitschr., 1909, p. 192.

Worker and Female.

The worker resembles the var. *foreli* more closely, especially in stature and in the length of the maxillary palpi. The female resembles the typical *exsecta,* especially in stature.

Switzerland; Vosges Mts.

The nests of this variety are described by Forel as being intermediate between those of *foreli* and the typical *exsecta.*

78. F. EXSECTA PRESSILABRIS var. RUFOMACULATA Ruzsky.

F. exsecta pressilabris var. *rufomaculata* Ruzsky, Arb. Ges. naturf. Kasan, 1895, **28**, p. 13, ☿; Berlin. ent. zeitschr., 1896, **41**, p. 68; Formicar. Imper. Ross., 1906, p. 369; Emery, Deutsch. ent. zeitschr., 1909, p. 192.

Worker. Characterized by having the base of the first gastric segment with a red spot at the base. Legs yellowish brown.

Southeastern Russia.

79. F. SUECICA Adlerz.

F. suecica Adlerz, Öfvers. Vet. Acad. förhandl., 1902, p. 263, ☿ ♀ ♂.
F. exsecta subsp. *suecica* Emery, Deutsch. ent. zeitschr., 1909, p. 193, ☿ ♀ ♂.

WORKER. 4–6.3 mm.

Closely related to *F. exsecta.* Head proportionally broader, with less deeply excised posterior border and more rounded posterior corners, the cheeks rather convex, front depressed. Mandibles 8-toothed, with 2 or 3 minute additional teeth on the basal border. Clypeus indistinctly carinate, its anterior portion flattened and its border slightly reflected. Frontal area triangular, a little longer than broad. Maxillary palpi long, 6-jointed. Epinotum in profile not so rounded as in *exsecta,* distinctly angular, with distinct base and declivity, the former horizontal, very feebly convex, the latter very sloping. Petiole very much like that of *exsecta,* its margin sharp and deeply excised.

Head and gaster rather smooth and shining, finely shagreened. Frontal area smooth and shining.

Pilosity and pubescence very feebly developed, only the clypeus, mouthparts, venter, and tip of gaster with sparse hairs.

Head, thorax, and petiole ferruginous; front, vertex, and posterior portion of head, especially in small workers, often darker; antennal funiculi, anterior border of clypeus, dental border of mandibles, sometimes also the legs in part, brown; gaster blackish brown.

FEMALE. Length 5–6.3 mm.

Very small. Resembling the worker in the structure of the head; the thorax and petiole as in *F. exsecta.*

Body very shining, especially the head, thoracic dorsum, and gaster.

Pilosity and pubescence as in the worker.

Upper surface of head, posterior border of pronotum, mesonotum, scutellum, and border of petiole and gaster, except the anal region, blackish brown. Remainder of body darker or lighter yellowish or reddish brown. Wings very pale grayish hyaline, with pale brown veins and darker stigma.

MALE. Length 6–6.5 mm.

More slender than the male *exsecta;* posterior border of head broadly excised but sometimes indistinctly so. Mandibles broad, pointed, edentulous. Palpi as in the worker. Petiole low and thick, as broad as high, truncated above, with blunt, entire border.

More shining than the male of *exsecta,* especially the gaster. Frontal area and mandibles more shining than the remainder of the head.

Hairs and pubescence no more abundant than in the worker and female, even sparser on the gaster. Mesonotum with a few short, erect hairs. Eyes hairless.

Black; genitalia and legs yellowish, middle portions of femora and tibiae more or less infuscated. Wings pale grayish hyaline as in the female.

Sweden: Alnö Island, in the Gulf of Bothnia, near Sundsvall.

This form is regarded by Emery as an extreme race of *F. exsecta,* but the differences of structure and habit seem to me to be sufficient to entitle it to the rank of a species, as it was originally described by Adlerz. According to this observer it does not build mound-nests like the typical *exsecta* and its subspecies and varieties but resembles *F. truncicola* in establishing its formicaries in rotten stumps or logs which it banks with fine vegetable detritus.

Fusca Group.

80. F. FUSCA FUSCA Linné.

F. fusca Linné, Syst. nat. ed. 10, 1758, **1**, p. 580; Fauna Suec., ed. 2, 1761, p. 426; deGeer, Mem. hist. ins., 1771, **2**, p. 1, pl. 42, figs. 12–15; Fabricius, Spec. ins., 1781, **1**, p. 490; Mantissa ins., 1787, **1**, p. 308; Olivier, Encycl. meth. insect., 1791, **1**, p. 493; Fabricius, Ent. syst., 1793, **2**, p. 352; Latreille, Essai hist. fourmis France, 1798, p. 39, ☿ ♀ ♂; Hist. nat. fourmis, 1802, p. 159, pl. 6, fig. 32, e ☿ ♀ ♂; Fabricius, Syst. Piez., 1804, p. 399; Latreille, Gen. Crust. ins. 1809, **4**, p. 126; Lepeletier, Hist. nat. insect. Hymén., 1836, **1**, p. 205, ☿ ♀ ♂; Westwood, Introd. mod. class. insects, 1840, **2**, Synop., p. 83; Nylander, Acta Soc. Fennica, 1846, **2**, p. 917, 919, ☿ ♀ ♂, pl. 18, fig. 23; Ibid., 1849, **3**, p. 27, 30; Förster, Stettin. ent. zeit., 1853, **14**, p. 141; F. Smith, Trans. Ent. soc. Lond., 1855, ser. 2, **3**, p. 104, ☿ ♀ ♂, pl. 9, fig. 15; Mayr, Verh. Zool. bot. ver. Wien, 1855, **5**, p. 346, ☿ ♀ ♂; Nylander Ann. sci. nat. Zool., 1856, ser. 4, **5**, p. 65, ☿ ♀ ♂; F. Smith, List Brit. anim. Brit. mus., 1858, pt. 6, p. 5, pl. 3, fig. 14; Meinert, Natur. abh. Dansk. vid. selsk., 1860, ser. 5, **5**, p. 44, ☿ ♀ ♂; Mayr, Europ. Formicid., 1861, p. 47–49, ☿ ♀ ♂; Taschenberg, Hymen. Deutschl., 1866, p. 239; Forel, Denks. Schweiz., gesell. naturw., 1874, **26**, p. 53, 56, 58, 356, ☿ ♀ ♂; Bull. Soc. Vaud. sci. nat., 1875, ser. 2, **14**, p. 60; Lubbock, Journ. Linn. soc. Zool., 1877, **13**, p. 217, pl. 17, fig. 3; Ern. André, Spec. Hymén. Europe, 1882, **2**, pt. 14, p. 182, 186, 190, pl. 5, fig. 12, ☿ ♀ ♂; Adlerz. Bih. Svensk. vet. akad. Handl., 1886, **11**, p. 286, 290, ☿ ♀ ♂; Wasmann, Deutsch. ent. zeitschr., 1887, **31**, p. 109; Dalla Torre, Catalog. Hymen., 1893, **7**, p. 196; Ruzsky, Formicar. Imper. Ross., 1905, p. 373, 1 fig.; Emery, Deutsch. ent. zeitschr., 1909, p. 193, ☿ ♀ ♂; Ibid., 1912, p. 672.

F. fusca var. *glacialis* Wheeler, Bull. Amer. mus. nat. hist., 1908, **24**, p. 624, ☿ ♀ ♂.

WORKER. Length 4–6.5 mm.

Body rather slender. Head longer than broad, narrower in front than behind, with straight posterior and lateral borders. Eyes large. Mandibles 8-toothed. Clypeus sharply carinate its entire length, with entire, rounded, projecting anterior border. Frontal carinae moderately diverging behind. Antennae rather slender, the scapes slightly thickened towards their tips, the basal joints of the funiculus scarcely longer and but little more slender than the penultimate joints. Maxillary palpi moderately long. Thorax narrow; pro- and mesonotum but slightly convex, rather depressed above; mesoëpinotal constriction shallow; epinotum with subequal base and declivity, both straight in profile, the former horizontal, the latter sloping, meeting at a distinct but obtuse angle. Petiole narrow, cuneate in profile, with convex anterior and flattened posterior surface, its border rather sharp, entire and broadly rounded when seen from behind. Gaster small, legs rather slender.

Subopaque and very finely and sharply shagreened; mandibles coarsely striatopunctate; clypeus finely longitudinally striate. Frontal area subopaque, only the sutures surrounding it shining.

Hairs and pubescence whitish, the former short, erect, very sparse, confined to the upper surface of the head, thorax and gaster, coxae and venter. Eyes hairless. Pubescence dense on the head, thorax, and gaster, longest on the gaster, giving the surface a slightly pruinose, but not a silky appearance.

Black; mandibles, scapes, basal joints of funiculi, and legs deep red, femora and tibiae, except the knees, often darker.

FEMALE. Length 7–10 mm.

Smaller than the females of *rufa,* but the gaster proportionally larger and more elliptical. Thorax broader than the head, which, excluding the mandibles, is as broad as long. Petiole compressed anteroposteriorly and broader than in the worker. Wings long.

Sculpture, pilosity, and color much as in the worker, except that the gaster is very smooth and shining, with much more dilute pubescence. Wings nearly colorless or very slightly yellowish; stigma brown.

MALE. Length 8–10 mm.

Body slender. Mandibles narrow, pointed, often, if not always, denticulate. Head broad behind and considerably narrowed in front, with large eyes, straight posterior border and cheeks and rounded posterior corners. Clypeus convex, bluntly carinate. Thorax broader than the head. Petiole transverse, somewhat compressed anteroposteriorly towards the superior border, which is blunt and, seen from behind, with a broad and very shallow median excision. Gaster rather long and narrow. Stipes but little longer than the volsellae and sagittae.

Head and thorax, including the mandibles and frontal area, subopaque. Gaster distinctly shining.

Hairs and pubescence much as in the worker, the former absent on the upper surface of the gaster. Eyes hairless.

Black; gaster often more brownish; scapes and tips of mandibles dark brown; legs and genitalia yellow. Bases of the coxae and sometimes also the last tarsal joint of each foot, black. Wings grayish hyaline, scarcely darker than in the female.

Widely distributed through North and Central Eurasia; but occurring only in mountainous country in Southern Europe and there often at considerable elevations (up to 2,400 meters in the Alps, according to Forel). This form is also widely distributed through Boreal America. I have examined specimens from the following localities: —

Newfoundland: Cod Roy and Bay of Islands (L. P. Gratacap); East Coast (W. T. Davis); Spruce Brook (Amer. Mus. Nat. Hist. Coll.).

Nova Scotia: Digby (J. Russell); Port Maitland (W. Reiff); Boisdale, Cape Breton (W. T. Davis).

New Brunswick: St. Stephen and St. Andrews (Centr. Exper. Farms Coll.).

Quebec: Point Comfort, James Bay (A. Skinner); Hull, Kingsmere (Wheeler).

Ontario: Rat Portage (J. C. Bradley); Guelph, Ottawa (Wheeler); Marshall's Bay near Arnprior (C. G. Hewitt).

Saskatchewan: Methy Lake (R. Kennicott).

British Columbia: Carbonate, on the Columbia River, 2,800 ft. (J. C. Bradley).

Maine: South Harpswell, Lower Goose Island, Casco Bay (Wheeler); Monmouth (Frost).

New Hampshire: Mt. Washington (Mrs. A. T. Slosson); Mt. Moosilauke, 1,500–3,000 ft. (Wheeler).

Vermont: Lyndon (A. L. Melander).

Massachusetts: Blue Hills, near Boston (Wheeler).

New York: Ramapo Mountains (Wheeler); Niagara Falls (Amer. Mus. Nat. Hist. Coll.).

North Carolina: Lake Toxaway (Mrs. A. T. Slosson); Black Mountains (W. Beutenmüller).

Michigan: Pequaming, Baraga County (M. Hebard); Isle Royale (H. A. Gleason).

Colorado: Cripple Creek, 10,200 ft. (Wheeler); Steamboat Springs (T. D. A. Cockerell).

Montana: Weeksville (S. Henshaw); Helena, Elkhorn, Nigger Hill, Powell County (W. M. Mann).

Idaho: Moscow (J. M. Aldrich).

Washington: Pullman (W. M. Mann).

On careful examination I am unable to detect any important differences between the form which I described as the var. *glacialis* from Maine and the true European *fusca*. The wings of the males and females in the American form are perhaps slightly darker, but the tint is variable in European specimens. The sculpture, color, and pubescence are identical in the two forms. The specimens from Newfoundland, including in all probability those from St. Pierre and Miquelon, Newfoundland, mentioned by Emery (Zool. jahrb. Syst., 1893, **7**, p. 660), and the specimens from Nova Scotia and New Brunswick agree very closely with the cotypes from Maine. The western forms are often a little more like *subsericea* in pubescence and may be regarded as transitional to that variety. Should it be possible on further study to detect any satisfactory differences between American and Eurasian specimens, the term *glacialis* would, of course, have to be reinstated.

The colonies of the American *fusca* are often much larger than those which I have seen in Europe. In both continents it nests under stones or logs or in rude craters or small earthen mounds. The workers are extremely timid. This timidity, which characterizes all the varieties and subspecies of *F. fusca*, together with its extreme fecundity, has made it an ideal host for a large number of the parasitic species of Formica of the *sanguinea, rufa, microgyna,* and *exsecta* groups.

81. F. FUSCA FUSCA var. GLEBARIA Nylander.

F. glebaria Nylander, Acta. Soc. Fennica, 1846, **2**, p. 917, ☿ ♀, taf. 18, fig. 23; Förster, Hymen. stud., 1850, **1**, p. 31, ☿ ♀ ♂.

F. fusca var. *glebaria* Emery, Deutsch. ent. zeitschr., 1909, p. 196, ☿ ♀; Karawajew, Rev. Russe ent., 1909, p. 268.

F. fusca subsp. *glebaria* Emery, Deutsch. ent. zeitschr., 1912, p. 672.

WORKER. Length 4–6.5 mm.

Differing from the typical *fusca* in color and pilosity. The body is deep brown or at any rate not deep black, and the pubescence is longer and more abundant, especially on the gaster, so that the body is distinctly silky. The front of the head, the sutures of the thorax, the scapes, and articulations of the legs are pale and more yellowish or reddish.

FEMALE. Length: 7–9 mm.

Resembling the worker in color and pilosity. The gaster is not smooth and shining as in the typical *fusca* but subopaque and covered with much denser pubescence and appearing glossy or silky. Wings distinctly infuscated at their bases.

MALE. Length 8–9 mm.

Apparently indistinguishable from the male of the typical form. Perhaps the pubescence on the gaster is a little longer and denser and this region therefore a little less shining. Wings as in the female.

Like *fusca* this variety is widely distributed through Eurasia. It has been introduced into gardens in Algiers. Emery states that it does not occur in the smaller southern islands in the Mediterranean and that it is absent from Crete. Krausse has recently taken it, however, in Sardinia. Unlike the true *fusca*, it prefers the lowlands and especially gardens and meadows, where it builds small mound-nests. If the ground is very dry the nests may be entirely subterranean like those of *F. rufibarbis*.

Emery has recently come to regard *glebaria* as a good subspecies, instead of as a mere variety of *fusca*, because he finds that the workers of the typical form of this species will not rear the pupae of *glebaria*. It is not at all clear that such behavior necessarily constitutes a criterion of the taxonomic status of a subspecies, since it will not hold even for species. Moreover, if *glebaria* is raised to subspecific rank it will be necessary to do the same with many of our American forms of *fusca*, such as *subsericea*, *neoclara*, *gelida*, etc., and I am not prepared to regard these as more than varieties.

82. F. FUSCA FUSCA var. RUBESCENS Forel.

F. fusca var. *rubescens* Forel, Bull. Soc. ent. Belg., 1904, **48**, p. 423, ☿ ; Emery, Deutsch. ent. zeitschr., 1909, p. 196, ☿ ♀.

WORKER. Length 4–6.5 mm.

Sculpture and pubescence as in *glebaria*. In the large worker the anterior portion of the head, the thorax, scapes, first funicular joint, and the legs are yellowish red, with the exception of two almost confluent fuscous spots resembling those of *F. pratensis*, on the pro- and mesonotum. The small workers are scarcely distinguishable from those of var. *glebaria*, the red color being very feebly developed or absent.

FEMALE. Length 7–9 mm.

Lower portions of thorax and the petiole more or less red, the color of the remainder of the body as in *glebaria*, the gaster subopaque and covered with short, silky pubescence. Wings distinctly infuscated at their bases.

MALE. Length 8–9 mm.

Indistinguishable by any reliable characters from the males of *fusca* and its var. *glebaria*.

This variety is known only from Central Europe. It is common in Switzerland, the type locality, inhabiting the same stations and nesting in the same manner as the var. *glebaria*. It is often confused with *F. rufibarbis* on account of its color, but this species usually lacks the dark spots on the thorax and is fierce and aggressive, whereas *glebaria*, like all the other varieties of *fusca*, is very timid.

83. F. FUSCA FUSCA var. JAPONICA Motschulsky.

F. japonica Motschulsky, Bull. Soc. nat. Moscou, 1866, **39**, p. 183, ☿.

F. fusca var. *nipponensis* Forel, Mitth. Schweiz. ent. gesell., 1900, **10**, p. 270, ☿; Mitth. Naturh. mus. Hamburg, 1901, **18**, p. 66, ♀; Ern. André, Bull. Mus. hist. nat. Paris, 1903, p. 128; Wheeler, Bull. Amer. mus. nat. hist., 1906, **22**, p. 323, ☿.

F. fusca var. *japonica* Emery, Deutsch. ent. zeitschr., 1909, p. 197, ☿ ♀.

WORKER. Length: 4–5.5 mm.
Head, thorax, and gaster opaque, rather coarsely shagreened. Mandibles coarsely striatopunctate. Legs slightly shining. Hairs and pubescence white, the former short, sparse, on the gaster stubby and obtuse, the pubescence very short, moderately dense and giving the surface a slightly pruinose appearance. Body black; mandibles, antennae, tarsi, sutures of thorax, and articulations of legs brown.
FEMALE. Pilosity, sculpture, and color as in the worker.

This ant appears to be common in Japan. Forel's specimens came from the Island of Yezo and from Tokio. I have seen specimens from Misaki, Kanagawa (1,700 ft.), Yamanaka, and Takakiyama. According to Emery, Ruzsky has recorded this variety also from Mongolia. It approaches the North American var. *subsericea* in some respects, but is peculiar in the dull opacity of the body.

84. F. FUSCA FUSCA var. SUBSERICEA Say.

F. subsericea Say, Bost. journ. nat. hist., 1836, **1**, p. 289, ☿ ♀; Ed. Leconte, 1859, **2**, p. 734, ☿ ♂, Dalla Torre, Catalog. Hymen., 1893, **7**, p. 213.

F. fusca Mayr, Verh. Zool. bot. ver. Wien, 1886, **37**, p. 426.

F. fusca var. *subsericea* Emery, Zool. jahrb. Syst., 1893, **7**, p. 659, ☿ ♀ ♂; Wheeler, Bull. Amer. mus. nat. hist., 1905, **21**, p. 401; Occas. papers Bost. soc. nat. hist., 1906, **7**, no. 7, p. 19.

WORKER. Length 4–7 mm.
Base of epinotum often slightly convex, longer than the sloping, slightly concave declivity. Head in the largest workers as broad

as long. Petiole in such workers often rather broad, with compressed, feebly notched superior border.

Sculpture of the body somewhat finer and more superficial than in the typical *fusca*, so that the body is a little more shining. Moreover, it often has a faint metallic luster.

Hairs and pubescence pale yellow or whitish, the hairs short, sparse and erect, as in the true *fusca*, the pubescence longer and denser, giving the body and especially the gaster a silky appearance. Legs and scapes with equally dense but shorter pubescence.

Black; mandibles, antennae, tarsi, and articulations of legs dull red or brown.

FEMALE. Length 8–10.5 mm.

Large and stout. Like the worker in sculpture, pilosity, and color. Pubescence on the gaster even longer and more conspicuous. Legs more reddish. Wings uniformly and rather deeply infuscated.

MALE. Length 9–10.5 mm.

Resembling the male of the typical *fusca*, larger and more robust, with the pubescence, especially on the gaster, much longer and more abundant and the wings deeply infuscated as in the female. The shagreening of the body is coarse, so that the surface is rather opaque and even the gaster is only very slightly shining.

TYPE LOCALITY.— Indiana.

Indiana: Camelton, Hammond, Vedensburg, Wyandotte, Vawter Park, Arlington, Pine, Culver, Tippecanoe Lake, Shoals, Bass Lake (W. S. Blatchley).

Illinois: Rockford (Wheeler).

Michigan: Ann Arbor (J. Dawson); Porcupine Mountains (O. McCreary); Isle Royale (H. A. Gleason).

Colorado: Manitou (Wheeler).

Arizona: Miller Canyon, Huachuca Mountains (Biederman).

Washington: Olympia (T. Kincaid).

Kansas: Lawrence.

Pennsylvania: White Haven (J. C. Bradley).

Georgia: Black Rock Mountain, Rabun County (J. C. Bradley).

North Carolina: Black Mountain (W. Beutenmüller).

Virginia: Ashland (J. F. McClendon).

New Jersey: Caldwell (E. T. Cresson); New Brunswick (J. B. Smith); Great Notch, Jamesburg, Ft. Lee, Lakehurst (Wheeler); Montclair (G. v. Krockow); Newark.

New York: Central Park, New York City; Saugerties, Bergen Beach (G. v. Krockow); Garrison-on-Hudson (T. D. A. Cockerell); Bronxville and Mosholu (Wheeler); Kiamesha (C. T. Brues); Ithaca (Cornell Univ. Coll.).

Connecticut: Suffield (Geo. Dimmock); Branford, Cheshire, Mt. Carmel, New Haven (H. L. Viereck); New Haven, Salisbury (W. E. Britton); Cromwell, Hartford (A. Forel); Winsted, Norfolk, Colebrook (Wheeler).

Rhode Island: Providence (Davis).

Massachusetts: Sherborn, Wellesley, Andover (A. P. Morse); Essex County, Mt. Tom, Springfield (G. B. King); Springfield (J. A. Allen); Arlington, Cambridge (Mus. Comp. Zoöl.); Readville, Woods Hole, Boston (Wheeler); Medford (W. H. Dall).

New Hampshire: Holderness (A. P. Morse); Canobie Lake, West Ossipie (G. B. King); Mt. Moosilauke, 1,700 ft. (Wheeler).

Vermont: Hyde Park.

Maine: South Harpswell, Sebascoegan Island, Casco Bay (Wheeler).

Nova Scotia: Digby (J. Russell).

Ontario: Toronto (R. J. Crew); Ottawa (Centr. Exper. Farms Coll.); Guelph, Port Stanley (W. H. Wright).

This is the most abundant Formica in temperate North America and one of the most abundant insects, next to *Lasius niger* var. *americanus,* at least in the Eastern United States. Its colonies, which are often rather large, nest in sunny places under stones or in low flat "beds," or mounds, often a meter or more in diameter. Owing to its great abundance, it is the favorite host of the Nearctic forms of the *sanguinea, rufa,* and *exsecta* groups. It is a very cowardly ant and rarely resents disturbance of its nests unless it happens to be acting as the "slave," or auxiliary of *sanguinea.* Although the pure form of *subsericea* may be readily recognized, there occur forms which in sculpture and pilosity connect it with the true *fusca* and with the varieties *subaenescens* and *argentea,* and the workers of such forms are not always easy to identify.

85. F. FUSCA FUSCA var. ARGENTEA Wheeler.

F. fusca var. *argentata* Wheeler, Amer. nat., 1902, **36**, p. 952, *nota* ☿; Ants, 1910, p. 570.

F. fusca var. *argentea*, nom. nov. Wheeler, Psyche, 1912, **19**, p. 90.

WORKER. Length 4–7 mm.

Closely related to the var. *subsericea* but differing in the somewhat more slender body, longer legs, in the character of the pubescence, and in color. The pubescence is more glistening white and denser, so that the whole body has a silvery luster. The body is dark reddish brown or brownish black, instead of black, the mandibles, corners of clypeus, anterior borders of cheeks, antennae, and legs light red or even yellowish. The last funicular joint, femora, and tibiae are often darker, except at the articulations. In some specimens the femora are blackish, with the knees, tibiae and tarsi reddish yellow.

FEMALE. Length 8–10.5 mm.

Resembling the worker in color and pubescence, except that the body is darker and less silvery. Differing from the female of all the preceding forms of *fusca* in having the wings colorless or very faintly tinged with yellow near the anterior border, veins yellow, stigma brown.

MALE. Length 9–10 mm.

Differing from the male of the preceding forms in having colorless wings and in the color of the body. The head and thorax are black or dark brown, the gaster sometimes paler. Legs, genitalia, antennae, and mandibles clear yellow.

TYPE LOCALITY.— Illinois: Rockford, (Wheeler).

Illinois: Algonquin (W. A. Nason); Galesburg (Centr. Exp. Farms Coll.).

Washington: Yakima River (S. Henshaw).

Oregon: Corvallis, The Dallas (Amer. Mus. Nat. Hist. Coll.).

California: Palo Alto (H. Heath); Corte Madera Creek (W. M. Mann); Harris, Humboldt Co. (J. C. Bradley).

Arizona: Coconino Forest, Grand Canyon, 7,000 ft., Williams, 7,000 ft. (Wheeler); Miller Canyon, Huachuca Mountains (H. A. Wenzel).

New Mexico: Gallinas Canyon, Pecos (T. D. A. Cockerell); Manzanares (Miss Mary Cooper); Las Vegas (Wheeler).

Montana: Helena (W. M. Mann).

Colorado: Colorado Springs, Colorado City, Florissant, Buena Vista, Salida, Pike's Peak, 10,000–11,000 ft. (Wheeler); Troublesome, Boulder (S. A. Rohwer); Salina (T. D. A. Cockerell).

South Dakota: Pierre (S. S. Visher).

Utah: East Mill Creek, Willow Canyon (R. V. Chamberlin).

Kansas: Lawrence.

Michigan: Porcupine Mountains (O. McCreary); Isle Royale (Gleason); Marquette (M. Downing).

New Hampshire: Durham, White Mountains (W. F. Fiske).

Massachusetts: Ellisville, Annisquam (Wheeler); Cotuit, Woods Hole (Miss A. M. Fielde).

British Columbia: Loon Lake, Spillimacheen River, Selkirk Mountains (J. C. Bradley).

This variety, which in its pure form is readily distinguished by the beautiful silvery pubescence and pale legs and antennae of the worker and the clear wings of the male and female, is very widely distributed. It evidently belongs to the colder portions of the transition zone and is common in the mountains of the western part of the country between elevations of 7,000 and 11,000 ft., but more sporadic in the Eastern States. It nests in the sand dunes and along the beaches of the New England coast but seems to be rather local.

86. F. FUSCA FUSCA var. MARCIDA, var. nov.

WORKER. Length 2.5–4.5 mm.

Closely allied to the typical *fusca* but averaging smaller. The sculpture and pubescence are much as in *fusca*. Body subopaque. Hairs very sparse and short. Body dark reddish brown, head and gaster blackish, sutures of thorax reddish or yellowish, mandibles, antennae, and legs pale yellowish brown, tips of funiculi and middle portions of femora somewhat darker.

FEMALE (DEÄLATED). Length 7–8 mm.

Like the female of the typical *fusca* but smaller; gaster and upper surface of thorax nearly as smooth and shining, with sparse pubescence. Body blackish brown, mandibles, legs, scapes, and bases of funiculi brownish yellow.

TYPE LOCALITY.— British Columbia: Prairie Hills, Selkirk Mountains (J. C. Bradley).

British Columbia: Howser, Golden, Carbonate, 2,600 ft. and Moraine Lake (J. C. Bradley); Golden (W. Wenman).

Alberta: Banff (N. B. Sanson).

Manitoba: Aweme (Jas. Fletcher).

Washington: Ellensburg, Kiona (W. M. Mann); Brinnon, Hood Canal (J. C. Bradley).

Described from nine deälated females and numerous workers. This variety, at first sight, resembles the European *glebaria*, but it is smaller and the female has a smooth, shining gaster and thoracic dorsum like the female of the typical *fusca*. The workers of some colonies are almost indistinguishable from the typical *fusca*, others are as clearly transitional to the varieties *gelida* and *argentea*.

A note by Mr. Bradley accompanying the specimens from Moraine Lake states that they "were gathered under a stone from which the snow had recently receded. The workers are quick and agile and hide under the stones and in moss. Quite a number of nests were found at about timber-line." These remarks indicate that *marcida* is an alpine variety like *gelida*.

87. F. FUSCA FUSCA var. SUBAENESCENS Emery.

F. fusca var. *subaenescens* Emery, Zool. jahrb. Syst., 1893, **7**, p. 659, ☿; Forel, Ann. Soc. ent. Belg., 1904, **48**, p. 153, ☿; Wheeler, Ants, 1910, p. 570.
?*F. fusca* var. *densiventris* Viereck, Trans. Amer. ent. soc., 1903, **29**, p. 73, ☿.

WORKER. Length: 4–7 mm.

Very closely related to the typical *fusca* but differing in having the body, and especially the gaster more shining. The gaster is finely shagreened and also finely punctate. The pubescence is much sparser than in *subsericea* and usually somewhat sparser than in the typical *fusca*, so that the surface is clearly visible. The hairs and pubescence are yellowish. Body black, with distinct bronzy reflections. Mandibles, antennae, and legs dark brown or dark red; tibiae and femora, except at the articulations, often darker.

FEMALE. Length 8–10 mm.

Resembling the worker in color, sculpture, and pilosity, but the gaster, posterior part of head and mesonotum even smoother and more shining. Wings rather deeply and uniformly infuscated, but slightly less than in the var. *subsericea*. Veins and stigma brown.

MALE. Length 8–10 mm.

Closely resembling the male of the typical *fusca*, but the wings somewhat more deeply infuscated and the gaster more shining and more sparsely and delicately pubescent. Head and thorax black, gaster dark brown; mandibles, legs, genitalia, and scapes clear yellow; funiculi light brown.

TYPE LOCALITY.— South Dakota: (Th. Pergande).

Utah: Little Willow Canyon (R. V. Chamberlin).

Colorado: Manitou, 7,000–8,000 ft., Colorado City and Pike's Peak, 10,000–11,000 ft. (Wheeler); Pike's Peak, printing office, 10,000 feet, Ward, 9,000 ft. (T. D. A. Cockerell).

New Mexico: Old Pecos Pueblo, Pecos, Top of Las Vegas Range, 11,000 ft. (T. D. A. Cockerell); Barela Mesa (Miss Anna Gohrman); Manzanares (Miss Mary Cooper); Harvey's Ranch, Las Vegas Range, 9,600 ft. (Miss Ruth Reynolds); Cloudcroft (H. Skinner).

Montana: Nigger Hill, Powell County (W. M. Mann).

California: King's River Canyon, 8,000 ft. (H. Heath); Alta Peak, Sequoia National Park, 9,500–11,000 ft. (J. C. Bradley).

Washington: San Juan Island (W. M. Mann); Brinnon, Hoods Canal (J. C. Bradley).

Idaho: Troy (W. M. Mann).

Maine: Lower Goose Island, Casco Bay (Wheeler).

New Hampshire: Mt. Washington (C. S. Bacon); Franconia (Mrs. A. T. Slosson).

Massachusetts: Wellesley (A. P. Morse); Blue Hills (Wheeler).
Connecticut: Colebrook (Wheeler).
New York: Ithaca (J. C. Bradley); Bedford (Wheeler).
Ontario: Guelph, Port Stanley (W. H. Wright).
Quebec: Kingsmere (Wheeler).
British Columbia: Howser, Carbonate, Selkirk Mts. (J. C. Bradley); Mt. Goodsir, 7,000 ft. (E. Whymper).
Alberta: Vermillion Pass, 5,000–6,500 ft. (E. Whymper).

This form differs considerably in the amount of pubescence on the gaster. The specimens from New Mexico, especially, have the pubescence nearly as dense and abundant as in *subsericea*, but as the ground surface is coppery and partially visible I have included them in this variety. They are, perhaps, the form described by Viereck as var. *densiventris*, but his original description based on two workers from Beulah, New Mexico (8,000 feet), is far from clear, and I have not been able to examine the types. Specimens from Rockford, Ill., agree very closely with Emery's description based on material from South Dakota and Connecticut. The worker specimens from Alta Peak, Calif., are very small and the pubescence is very delicate. They are decidedly bronzy, but in other respects might be referred to the typical *fusca*.

F. subaenescens nests under stones in cold, shady woods. Like the var. *argentea* it is rare and sporadic at lower altitudes and latitudes in the transitional zone and is evidently a boreal form, slightly more eurythermal than the true *fusca*.

88. F. FUSCA FUSCA var. GELIDA, var. nov.

F. fusca var. *neorufibarbis* Forel, Ann. Soc. ent. Belg., 1904, **48**, p. 153, ☿ ♀; Pergande, Proc. Wash. acad. sci., 1900, **2**, p. 519; Wheeler, Bull. Amer. mus. nat. hist., 1906, **22**, p. 344; Ants, 1910, p. 570.

Worker. Length 2.5–5 mm.

Head and thorax subopaque, frontal area and gaster shining and rather smooth. Hairs as in the typical *fusca*, pale yellow; pubescence much sparser, not only on the gaster but also on the head and thorax, so that the ground surface of the body is fully revealed. This is rather densely and sharply shagreened on the head and thorax, but very finely shagreened and sparsely punctate on the gaster.

Reddish brown, posterior half of head above black, sometimes with a bronzy reflection. Gaster often as dark as the top of the head. Thorax more or less infuscated. In large workers the infuscation is often confined to a spot on the pronotum and one on the mesonotum; medium sized workers often have the pleurae more or less infuscated and in the smallest workers the whole thorax may be dark brown. Base of gaster and venter usually paler than the upper surface. Petiole more compressed anteroposteriorly, with flatter anterior and posterior surfaces and sharper border than in any of the preceding forms of *fusca*.

Female. Length 6–8 mm.

Resembling the worker, but the gaster even more shining. This region is also more spherical and less elliptical than in the other forms of *fusca*. The head and thorax are subopaque, except the frontal area, which is shining. Posterior border of the pronotum and the disc of the mesonotum with a few large, scattered punctures.

Reddish brown, posterior portion of head, upper surface of gaster, posterior border of pronotum, the mesonotum, scutellum, and metanotum blackish or dark brown. The pleurae may also be clouded with this color. Petiole and legs more yellowish brown. In some specimens (from California) the thorax is pure reddish brown, with three large spots on the mesonotum, the metanotum and posterior portion of the scutellum black. Wings colorless, with pale brown veins and darker stigma.

Male. Length 6–7 mm.

Head and thorax, including the frontal area, opaque; mesonotum covered with coarse, scattered punctures. Epinotum, petiole, and gaster shining. Erect hairs on thoracic dorsum, petiole, and base of gaster rather abundant. Pubescence very sparse and rather long. Black; gaster dark brown; genital appendages distinctly infuscated. Legs yellow, middle portions of femora slightly infuscated. Antennae black; only the tips of the mandibles brownish. Wings as in the female.

Type locality.— Colorado: Ward, 9,000 ft. (T. D. A. Cockerell).

Colorado: Arapahoe Peak, timberline, Long's Peak, 12,500 ft. (T. D. A. Cockerell); Cripple Creek, 10,200 ft., Cheyenne Mountain, 8,000 ft. (Wheeler); Canyon City (P. J. Schmitt).

New Mexico: N. E. Truches Peak, 12,000–13,000 ft., above timberline (Mrs. W. P. Cockerell and Miss Ada Springer); Harvey's Ranch, Las Vegas Range, 9,600 ft. (Miss Ruth Reynolds); Top of Las Vegas Range, 11,000 ft. (T. D. A. Cockerell).

Arizona: Coconino Forest, Grand Canyon, 7,000 ft. (Wheeler).

California: Alta Peak, Sequoia National Park, 9,500–11,000 ft., Blue Lake, Humboldt Co. (J. C. Bradley).

Oregon: (Amer. Mus. Nat. Hist. Coll.).

Washington: Three Brothers, Olympic Range (J. C. Bradley).

Michigan: Porcupine Mountains, Isle Royale (O. McCreary).

New Hampshire: Mt. Washington, 3,840 ft. (W. Reiff); Lafayette, 4,000 ft., Sphagnum bog (J. H. Emerton).

Alaska: Kassiloff Lake, Kenai Peninsula (Berlin Mus.); Sitka, Metlakaktla, Kadiak (Th. Pergande); Homer (A. Mehner).

British Columbia: Vancouver (A. L. Melander); Lake Louise, Hecto, Prairie Hills, Selkirk Mountains, above timberline, Fielde, Roger's Pass (J. C. Bradley).

Alberta: Laggan, Banff (J. C. Bradley); Emerald Summit Lake, Vermillion Pass, Vermillion Valley, 6,100 ft., Yoho Valley 4,600 ft., Ice River Valley 5,000 ft. (E. Whymper).

Saskatchewan: Methy Lake (R. Kennicott).

Ontario: Rat Portage (J. C. Bradley).

Quebec: Saguenay River (Geo. Engelhardt); Anticosti Island (S. Henshaw); East Maine River (A. Skinner); Mingan Island, Niapisca Island (S. Henshaw).

Labrador: Square Island (A. S. Packard); St. Lewis Inlet.

Newfoundland: Bay of Islands (L. P. Gratacap); Spruce Brook, Port Saunders, Port au Croix (Amer. Mus. Nat. Hist. Coll.).

Nova Scotia: Digby (J. Russell).

This variety has been confounded by Forel, Pergande, and myself, and possibly also by Emery, with the true *neorufibarbis* described below. It is, however, a perfectly distinct form, which, notwithstanding its wide distribution as shown by the preceding list of localities, is the most stenothermal and alpine of all our American forms of *fusca*. It is closely related to *subaenescens* in sculpture and pubescence but is characterized by the deep red color of the thorax and the constant infuscation of the pro- and mesonotum even in large specimens. I have found it nesting in rather small colonies under stones or in logs in woods or shady canyons at high altitudes, just below timberline.

89. F. FUSCA FUSCA var. NEORUFIBARBIS Emery.

F. fusca var. *neorufibarbis* Emery, Zool. jahrb. Syst., 1894, **7**, p. 660, ☿ .

WORKER. Length 3–6 mm.

Like the variety *gelida* in sculpture. Frontal area shining. Head and thorax subopaque, gaster shining, transversely shagreened. Hairs yellow, very short and sparse, absent on the thorax. Pubescence finer and a little denser on the gaster than in *neorufibarbis* but not concealing the shining surface. Head black, cheeks, front, clypeus, and mandibles dark brown. Thorax, petiole, legs, scapes, and base of funiculi clear yellowish red, the legs a little paler; only in small workers is there a slight tendency to infuscation of the thoracic dorsum. Gaster black or dark reddish brown, venter and base of first segment often paler. Petiole as in *neorufibarbis.*

FEMALE (DEÄLATED). Length 7 mm.

Very closely resembling the female of *gelida,* especially the form with pale thorax, having the mesonotum ornamented with three large dark brown or black blotches, but the pubescence, especially on the gaster, is finer and denser. Frontal area shining.

TYPE LOCALITY.— South Dakota: Hill City (Th. Pergande).

South Dakota: Harding County (S. S. Visher).

Utah: Willow Canyon, Salt Lake County, (R. V. Chamberlin).

Montana: Helena, Elkhorn, Nigger Hill, Powell County (W. M. Mann).

Idaho: Moscow (J. M. Aldrich).

Oregon: Portland (Amer. Mus. Nat. Hist. Coll.).

Washington: San Juan Island (W. M. Mann); North Bend (T. Kincaid); Union City (J. C. Bradley).

British Columbia: Alert Bay, Vancouver Island (H. I. Smith); Chillimack Valley (J. M. MaCoun); Carbonate, Fielde (J. C. Bradley).

Alberta: Lake Minnewonka (J. C. Bradley).

This variety, of which I have seen many workers, but only one female, is very closely related to the var. *gelida* though evidently occurring at much lower altitudes. Superficially it resembles the European *F. rufibarbis* but can be at once distinguished by its shining and much less pubescent gaster, smooth frontal area, and much sparser pilosity.

I believe that I am right in limiting Emery's name *neorufibarbis* to this form, first, because he describes the color as like that of the European *rufibarbis* and second, because he cites South Dakota and Nebraska among the list of localities. The form I have called *gelida* cannot occur in these states. Third, I possess two workers from South Dakota sent me several years ago by Pergande under the name "*neorufibarbis.*" These evidently belonged to the cotype series. If Emery actually included both forms under the latter name, it should be applied to the specimens from South Dakota, the first locality mentioned, and not to the specimens from Colorado, Montana, and California, which in part at least were probably referable to *gelida.*

90. F. FUSCA FUSCA var. NEOCLARA Emery.

F. fusca var. *neoclara* Emery, Zool. jahrb. Syst., 1894, **7**, p. 661, ☿; Wheeler, Ants, 1910, p. 570.

F. cinereorufibarbis Marsh, Bull. 64, Bur. ent. U. S. dept. agric. pt. 9, p. 73.

WORKER. Length 3–6 mm.

Epinotum rather rounded, not angular in profile. Petiole with a small notch in the transverse superior border; seen from behind cordate. Body opaque, shagreened; gaster only slightly lustrous; frontal area not smooth or shining. Hairs yellow, very short, sparse. Pubescence dense and rather long, especially on the head and gaster, where it conceals the surface. Body and appendages pale red; mandibles darker; vertex, tips of antennae, funiculi, and upper surface of gaster infuscated.

FEMALE. Length 7–8 mm.

Resembling the worker in color, sculpture, and pubescence. The ocellar triangle, the gaster behind the first segment, the tips of the funiculi, the scutellum, and metanotum, more or less infuscated. Remainder of body and appendages pale yellowish red. In some specimens there are three elongate brown blotches on the mesonotum, and the posterodorsal portion of the head is infuscated. Pubescence, especially on the gaster, much longer than in the worker so that this region has a bright pruinose appearance. Wings colorless, with brown veins and stigma.

MALE. Length 7–8 mm.

In form resembling the male of the typical *fusca*. Mandibles narrow, edentate. Thorax robust, gaster slender. Petiole thick, low, transverse, with very blunt, feebly excised border. Opaque; gaster more shining. Pubescence grayish, long and dense. Black; gaster dark brown, mesonotum sometimes with a pair of yellow spots. Antennae dark brown. Genital appendages distinctly infuscated. Legs pale yellow. Wings as in the female.

TYPE LOCALITY.— Colorado.

Colorado: Boulder, Canyon City, Longmont (P. J. Schmitt); Colorado Springs, Colorado City, Salida, Denver (Wheeler); South Boulder, Salina, Modern (T. D. A. Cockerell); Greeley (J. M. Aldrich); Rocky Ford (H. O. Marsh).

New Mexico: Pecos and Las Vegas, 6,400 ft. (T. D. A. Cockerell).

This beautiful and easily recognized variety occurs in Colorado only at altitudes below 7,000 feet, usually nesting in the sandy soil of river valleys. It is conspicuously common in the streets and parks of Colorado Springs, where it makes flat mounds consisting of numerous small, more or less confluent craters not unlike the nests of *F. subsericea* in grassy places in the Eastern States, or of *F. cinerea* in sandy river valleys of Southern Europe. It is readily enslaved by the forms of *sanguinea* inhabiting the same region. The males and winged females mature during the latter half of July.

Mr. Geo. B. King has recorded the variety *neoclara* from Essex County, Mass., and I possess eight specimens bearing labels with this locality. They resemble Colorado specimens very closely, except that the petiole is not emarginate and therefore not cordate when seen from behind, and the gaster is not infuscated. These specimens may represent a distinct variety. If they are really specimens of *neoclara,* I am unable to account for their occurrence in Massachusetts unless they were accidentally introduced from the West. Of course, the locality labels may be erroneous.

91. F. FUSCA FUSCA var. BLANDA, var. nov.

WORKER. Length 3–3.5 mm.

Resembling *neoclara,* but smaller, with the whole body reddish brown, the legs, antennae, and mandibles paler, the head, gaster, and tips of funiculi not infuscated. Subopaque and very densely and finely punctate or shagreened, the head and gaster slightly shining. Frontal area opaque. Hairs and pubescence white, the former short and very sparse, absent on the thorax, the latter very fine and short, rather dense on the gaster, shorter on the head and thorax. Thorax with very feeble mesoëpinotal constriction, epinotum rather long, obtusely angular in profile, its base longer than the declivity which is very sloping. Petiole rather thick and blunt, with entire and rounded superior border.

Described from a dozen workers taken by Prof. Trevor Kincaid at Olympia, Washington (type locality), six workers taken by the same collector at Seattle, Wash., two workers taken by Mr. J. C. Bradley in the Yosemite Valley and four workers taken by the same collector at Lemon Cove, Tulare County, California. The status of this variety is somewhat problematical. It may be merely a very pale form of the var. *marcida,* although there is little variation in the series of workers examined. They differ from the worker *neoclara* in the uniform brown color of the body, the shorter and more delicate pubescence, the absence of a notch in the petiolar border, the narrower head and smaller size.

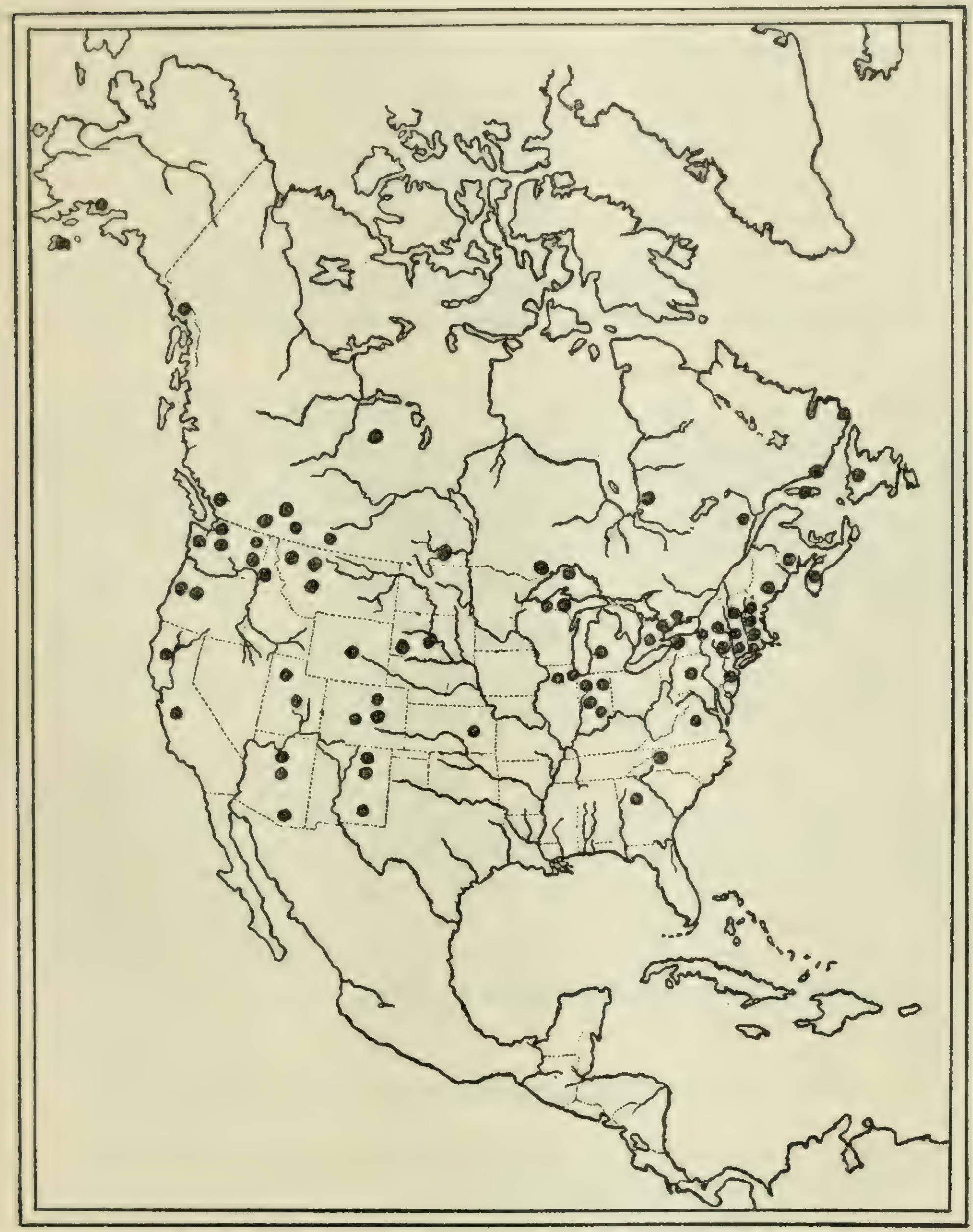

FIG. 6.— Distribution of the Nearctic forms of *Formica fusca*.

92. F. FUSCA PICEA Nylander.

F. picea Nylander, Acta Soc. Fennica, 1846, **2**, p. 917, 1059, ☿ ♀; Förster, Hymen. stud., 1850, **1**, p. 30, ☿.
F. gagates Meinert, Naturv. abh. Dansk. vid. selsk., 1860, ser. 5, **5**, p. 316, ☿ (*nec* ♀ ♂); Ruzsky, Formicar. Imper. Ross., 1905, p. 378; Dalla Torre, Catalog. Hymen., 1893, **7**, p. 198.
F. glabra White, Ants and their ways, 1883, p. 253.
F. transkaukasica Nassonov, Arb. Lab. zool. Univ. Moskau, 1889, **4**, p. 21.
F. fusca transkaukasica Ruzsky, Formicar. Imper. Ross., 1905, p. 384, ☿.
F. fusca gagates var. *filchneri* Forel, Ann. Soc. ent. Belg., 1907, **51**, p. 208, ☿.
F. fusca picea Emery, Deutsch. ent. zeitschr., 1909, p. 195, fig. 8, ☿.

WORKER. Length 3–6.5 mm.

About the size of the typical *fusca*. Epinotum angular in profile. Petiole rather broad, compressed anteroposteriorly, with sharp, entire border. Surface of body, including the frontal area, smooth and shining. Mandibles more opaque, finely striated and coarsely punctate. Hairs and pubescence very sparse. Color jet black; mandibles, antennae and legs dark red or dark brown; femora, tibiae, and ends of funiculi often darker.

FEMALE and MALE, judging from the descriptions, very similar to the corresponding phases of the typical *fusca*, but with the body of the female smooth and shining as in the worker.

Northern Europe and Asia to Eastern Siberia. According to Emery, this form represents the true *gagates* in Sweden, Finland, Russia, Eastern Siberia, and China, and has frequently been confounded with that species. He calls attention to the fact that oriental specimens sometimes have a few erect hairs on the underside of the head. I find these hairs in one of two workers of *picea* from Lahoul, Thibet, given me by Professor Forel.

93. F. FUSCA PICEA var. GAGATOIDES Ruzsky.

F. fusca var. *gagatoides* Ruzsky, Nachr. Russ. geogr. gesell., 1904, p. 289, ☿ ♀; Formicar. Imper. Ross., 1905, p. 377.
F. fusca picea var. *gagatoides* Emery, Deutsch. ent. zeitschr., 1909, p. 195, ☿ ♀.

WORKER and FEMALE.

Intermediate in its characters between *picea* and *fusca*.

Northern Europe.

94. F. GAGATES Latreille.

F. gagates Latreille, Essai hist. fourmis France, 1798, p. 36, ☿ ♀; Hist. nat. fourmis, 1802, p. 138, pl. 5, fig. 26, ☿ ♀; Lepeletier, Hist. nat. insect. Hymén., 1836, **1**, p. 200, ☿ ♀; Mayr, Verh. Zool. bot. ver. Wien, 1855, **5**, p. 347, ☿ ♀ ♂; Europ. Formicid., 1861, p. 46, ☿ ♀ ♂; Ern. André, Spec. Hymén. Europ., 1882, **2**, pt. 14, p. 182, 189, ☿ ♀ ♂; Dalla Torre, Catalog. Hymen., 1893, **7**, p. 198.

F. fusca st. *gagates* Forel, Denks. Schweiz. gesell. Naturw., 1874, **26**, p. 53, 217, ☿ ♀ ♂.

F. fusca gagates var. *muralewiczi* Ruzsky, Formicar. Imper Ross., 1905, p. 384.

F. fusca subsp. *gagates* Emery, Deutsch. ent. zeitschr., 1909, p. 194, fig. 7, ☿ ♀ ♂.

WORKER. Length 5–7.5 mm.

Closely related to *F. fusca*. Large and robust. Epinotum in profile rounded, without an angle between the base and declivity. Petiole broad, much compressed anteroposteriorly, with thin, sharp border.

Nearly the whole body smooth and shining, usually also the frontal area. Gaster very shining, very finely and transversely shagreened.

Hairs whitish, coarse, rather abundant on the gaster but very sparse elsewhere. Pubescence short and sparse, not concealing the shining surface.

Body deep black, mandibles dark brown, antennae, except their tips and legs, dark red or brown, with middle portion of the femora and tibiae sometimes black.

FEMALE. Length 9–11 mm.

Body robust. Head and thorax slightly, gaster more shining; Mesonotum with a few scattered foveolae. Pubescence on gaster very sparse. Color like that of the worker. Wings usually deeply and uniformly infuscated.

MALE. Length 9–10 mm.

Closely resembling the male of the typical *fusca* in color and sculpture but the pubescence longer and more abundant so that the body has a silky luster. Hairs almost absent, except on the venter. Petiole thick, with very blunt, entire or nearly entire superior border.

This form, which I would regard as an independent species and not as a subspecies of *fusca*, is confined to Asia Minor and Southern Europe (Southern France, Italy, Southern Germany, Austro-Hungary, the Balkan Peninsula, and the Crimea). According to Emery, Mayr detected transitions between this form and *F. fusca picea* in material from the Caucasus. Forel, who has studied the habits of *gagates* in Austria and Canton Ticino, Switzerland, found it nesting in oak forests under large stones and roots. The galleries are large and deep.

It attends aphids on the oaks. Mayr has also noticed the association of *gagates* with these trees, a relation similar to that obtaining between the oaks and the species of Liometopum and the varieties and subspecies of *Camponotus fallax* in both hemispheres.

95. F. GAGATES var. FUSCO-GAGATES Forel.

?*F. gagates* var. *morio* Latreille, Essai hist. fourmis France, 1798, p. 36, ☿; Hist. nat. fourmis, 1802, p. 140.

F. fusco-gagates Forel, Denks. Schweiz. gesell. naturw., 1874, **26**, p. 54, ☿.

F. gagates var. *fusco-gagates* Dalla Torre, Catalog. Hymen., 1893, **7**, p. 199.

F. fusca gagates var. *fusco-gagates* Forel, Ann. Mus. St. Petersbourg, 1904, **8**, p. 384; Emery, Deutsch. ent. zeitschr., 1909, p. 195, ☿.

WORKER.

Intermediate between *fusca* and *gagates*, but smaller, the structure of the epinotum subangular in profile and therefore more as in *fusca*. Head, thorax, and frontal area more opaque than in *gagates*.

Italian Switzerland.

Emery believes that this form may be a hybrid.

96. F. RUFIBARBIS Fabricius.

F. rufa Fourcrois, Ent. Paris, 1758, **2**, p. 452 (*nec.* Linné).

F. pratensis Olivier, Encycl. meth. insect., 1791, **6** (*nec.* Retzius).

F. rufibarbis Fabricius, Ent. syst., 1793, **2**, p. 355, ☿; Syst. Piez., 1804, p. 402; Jurine, Nouv. meth. class. Hymén., 1807, p. 273, ☿ ♀ ♂; Ern. André, Rev. mag. zool., 1874, ser. 3, **2**, p. 185; Spec. Hymén. Europ., 1822, **2**, p. 182, 186, 189, pl. 2, fig. 7, ☿ ♀ ♂; Mayr, Fedtschenko's Turkestan. Formicid., 1877, p. 7; Lubbock, Ants, bees, wasps, ed. 5, 1882, p. 80; Forel, Bull. Soc. Vaud. sci. nat., 1884, ser. 2, **20**, p. 379; Dalla Torre, Catalog. Hymen., 1893, **7**, p. 209; Ruzsky, Formicar. Imper. Ross., 1905, p. 385, ☿ ♀ ♂.

F. cunicularia Latreille, Essai hist. fourmis France, 1798, p. 38, ☿ ♀ ♂; p. 40, ☿ ♀ ♂; Brullé, Exped. sci. Morée zool., 1832, **2**, p. 326; Lepeletier, Hist. nat. insect. Hymén., 1836, **1**, p. 203; Nylander, Acta Soc. Fennica, 1846, **2**, p. 913, 1059, pl. 18, fig. 17–19, ☿ ♀ ♂; Förster, Hymen. stud., 1850, **1**, p. 25, ☿ ♀ ♂; F. Smith, Trans. Ent. soc. Lond., 1855, ser. 2, **3**, p. 103, pl. 9, fig. 14, ☿ ♀ ♂; Mayr, Verh. Zool. bot. ver. Wien, 1855, **5**, p. 342, ☿ ♀ ♂; Europ. Formicid., 1861, p. 47.

F. fusca st. *rufibarbis* Forel, Denks. Schweiz. gesell. naturw., 1874, **26**, p. 53, ☿ ♀ ♂.

F. fusca subsp. *rufibarbis* Emery, Deutsch. ent. zeitschr., 1909, p. 197; Karawajew, Rev. Russ. ent., 1909, p. 269.

Worker. Length 4–7.5 mm.

Very closely related to *F. fusca* and scarcely differing in structural characters. The epinotum is distinctly angular in profile; the petiole rather broad, compressed anteroposteriorly, with broadly rounded, entire, rather sharp superior border. Legs and antennae long.

Head, thorax, and gaster, including the frontal area, opaque, densely shagreened, venter and legs feebly shining.

Hairs yellow, very sparse, present on the upper surface of the head, pronotum and gaster, and sometimes also on the petiole and other parts of the thorax. Pubescence dense and rather long, concealing the surface, but without a silky gloss.

Pale red, posterodorsal portion of head and the gaster blackish brown. Mandibles dark red; funiculi, except at the base, the coxae and sometimes also the middle portions of the femora, infuscated. Rarely in large workers the pro- and mesonotum are slightly infuscated. In small workers the infuscation of the thorax may be more extensive.

Female. Length 9–11 mm.

Resembling the worker in sculpture, pubescence, and color, but the posterior margin of the pronotum, the scutellum, metanotum, more or less of the pleural region, and three elongate blotches on the mesonotum, dark brown. The venter is usually reddish. Wings grayish hyaline, with pale brown veins and darker stigma. Hairs longer and more abundant than in the worker, especially on the thoracic dorsum.

Male. Length 9–10 mm.

Very similar to the male of *fusca* and its varieties, but the thorax and gaster are more robust. Mandibles pointed, edentate. Petiole though thick and low, with a sharp, compressed and very broadly and distinctly excised border.

Body opaque, gaster scarcely shining. Frontal area opaque.

Hairs very sparse, present on the head, thoracic dorsum, and venter, but absent elsewhere. Pubescence grayish, short and dense.

Black; legs and genitalia yellow; tarsi of the former and tips of appendages of the latter infuscated; tips of mandibles brown. Wings uniformly gray as in the female.

Widely distributed through Europe and Northern Asia and occurring in Sardinia though absent from the smaller Mediterranean Islands. It is a distinctly xerothermal form and in the Alps does not reach such an elevation as the typical *fusca*. According to Ruzsky, however, it occurs at an altitude of 3,000 m. in the Caucasus, and according to Forel even higher in the Himalayas.

Although this ant is so very close to *fusca* in its morphological structure, I have nevertheless followed the majority of authors in regarding it as a species and not as a subspecies of *fusca*. P. Huber and Forel long ago showed that its habits and disposition are peculiar. It nests under stones or in open ground without building craters or mounds and in disposition is quite unlike the *fusca* forms, being very agile, fierce, and aggressive. It also has a peculiar odor quite unlike that of *fusca*, and this, together with the pugnacious disposition, also characterizes the American varieties.

97. F. RUFIBARBIS var. GLAUCA Ruzsky.

F. rufibarbis var. *glauca* Ruzsky, Arb. Ges. naturf. Kasan, 1895, **28**, p. 20, ☿; Berlin. ent. zeitschr., 1896, **41**, p. 70; Forel, Ann. Mus. St. Petersbourg, 1904, **8**, p. 385; Ruzsky, Formicar. Imper. Ross. 1905, p. 396.

F. fusca rufibarbis var. *glauca* Emery, Deutsch. ent. zeitschr., 1909, p. 198, ☿.

WORKER. Differing from the typical form in having the pubescence denser and with a bluish, silky luster on the gaster. The color and pilosity are like those of the typical form.

Southern Russia and Western Siberia.

98. F. RUFIBARBIS var. SUBPILOSA Ruzsky.

F. rubibarbis var. *subpilosa* Ruzsky, Ants envir. Aral Sea (Russian), 1902, p. 9, ☿; Zool. jahrb. Syst., 1902, **17**, p. 472; Forel, Ann. Mus. St. Petersbourg, 1904, **8**, p. 18; Ruzsky, Formicar. Imper. Ross., 1905, p. 397; Karawajew, Hor. Soc. ent. Ross., 1909, **39**, p. 16.

F. fusca rufibarbis var. *subpilosa* Emery, Deutsch. ent. zeitschr., 1909, p. 198, ☿.

WORKER. Color as in pale specimens of the typical *rufibarbis*. Pubescence whitish, dense on the gaster, which therefore has a gray tinge. Hairs also whitish, short, covering the whole body, but absent on the gula. Petiole moderately thick, with rather sharp superior border.

Central Europe, Southern Russia, and Central Asia to Western China.

This form resembles *F. cinerea* var. *imitans* but can be distinguished by the absence of erect hairs on the gula. The color, according to Ruzsky, is very variable, specimens from the Aral Sea region being very pale, like the var. *clara*, whereas those from Central Europe, the Crimea, Southeastern Russia, and the Caucasus are darker and more like the typical *rufibarbis*.

99. F. RUFIBARBIS var. CLARA Forel.

F. rufibarbis var. *clara* Forel, Ann. Soc. ent. Belg., 1886, **30**, p. 206, ☿; Ann. Mus. St. Petersbourg, 1904, **8**, p. 384.

F. rufibarbis clara Ruzsky, Zool. jahrb. Syst., 1902, **17**, p. 471, ☿ ♀ ♂; Formicar. Imper. Ross., 1905, p. 399.

F. fusca rufibarbis var. *clara* Emery, Deutsch. ent. zeitschr., 1909, p. 198, ☿ ♀ ♂.

WORKER. Length 5–5.6 mm.

Head, thorax, petiole, legs, and scapes pale ferruginous red; gaster dark brown; mandibles reddish brown. Some specimens have the tips of the scapes brownish and a spot of the same color on the vertex and occiput. Erect hairs lacking or very sparse; pubescence delicate and not very dense.

FEMALE. Length 8–9.5 mm.

Of the same color as very pale forms of the typical *rufibarbis* or even paler. Pubescence dense, sericeous on the body. Erect hairs sparse.

MALE. Length 8.5–9 mm.

Head dark brown, thorax and gaster brownish yellow. Mesonotum with three dark brown blotches and with the scutellum, part of the epinotum, petiolar border and spots on the pleurae of the same color. Legs, mandibles, and scapes yellowish brown; funiculi brown; wings infuscated.

Southern Russia, Siberia, Central Asia, Caucasus, Syria.

100. F. RUFIBARBIS var. CAUCASICA Ruzsky.

F. rufibarbis clara var. *caucasica* Ruzsky, Formicar. Imper. Ross., 1905, p. 401, ☿.

F. fusca rufibarbis var. *caucasica* Emery, Deutsch. ent. zeitschr., 1909, p. 198, ☿.

WORKER. Color as in the var. *clara* or only slightly darker, red or brownish red. Erect hairs lacking, with sparse, appressed hairs only on the sides of the head and thorax and on the legs and clypeus. Sculpture of body more delicate than in the var. *clara*, the whole body being subopaque, with feeble luster. Frontal area smooth and shining.

Caucasus Mountains.

101. F. RUFIBARBIS var. OCCIDUA Wheeler.

F. rufibarbis Wheeler, Amer. nat., 1902, **36**, p. 947 *et seq*, ☿.

F. rufibarbis var. *occidentalis* Wheeler, Ants, 1910, p. 570.

F. rufibarbis var. *occidua*, nom. nov. Wheeler, Psyche, 1912, **19**, p. 90.

WORKER. Length 4–7.5 mm.

Differing from the typical form of Europe only in pilosity. The thorax is either entirely without erect hairs or has only a few on the pronotum. The pubescence on the gaster is usually somewhat denser and more silvery so that it has a grayish or glaucous tinge, somewhat as in the var. *glauca.*

Female. Length 10–11 mm.

Indistinguishable from paler colored females of the typical form. Hairs, especially on the thorax, much more abundant than in the worker.

Type locality.— California: Palo Alto, (H. Heath and W. M. Mann).

California: Pasadena, San Ysidro near Santa Barbara, Palmer's Canyon, San Gabriel Mountains (Wheeler); Mount Wilson, Three Rivers, Sissons, Berkeley, Wild Cat Canyon near San Pablo, Lemon Cove, Tulare County (J. C. Bradley); Los Angeles (F. Grinnell, Jr.); San José (H. Heath); Santa Cruz Island (R. V. Chamberlin).

Washington: Wawawai (W. M. Mann).

The specimens from Washington have somewhat smoother bodies, the hairs are completely absent on the thorax in all specimens and this region is spotted with black and the black of the posterior part of the head runs under onto the posterior part of the gula, so that these specimens may, perhaps, represent a distinct variety.

The habits of the Californian form are very similar to those of the European type. It is fierce and aggressive and nests under stones in the open live-oak groves on the warm slopes of the Coast Range, at rather low altitudes.

102. F. RUFIBARBIS var. GNAVA Buckley.

F. gnava Buckley, Proc. Ent. soc. Phil., 1866, **6**, p. 156, ☿ ♀ ♂; Dalla Torre, Catalog. Hymen., 1893, **7**, p. 199.

F. fusca var. *gnava* Wheeler, Trans. Tex. acad. sci., 1902, **4**, p. 19; Bull. Amer. mus. nat. hist., 1906, **22**, p. 344; Ants, 1910, p. 570.

F. fusca var. *subsericeo-neorufibarbis* (Emery) Wheeler, Trans. Tex. acad. sci., 1902, **4**, p. 19.

Worker. Length 3.5–6 mm.

Differing from the preceding variety and the typical form in its average smaller size and in the more finely shagreened and therefore more shining surface of the body. Frontal area opaque. The head and gaster, especially, are more shining than in any of the other varieties of *rufibarbis.* This is due in part to the finer and shorter pubescence. The head, thorax, petiole, and legs vary from light to dark brownish red or brown, with the top of the head and often also the pro-

and mesonotum infuscated. Tips of funiculi not infuscated. The gaster is black, not paler ventrally. Small workers often have the head and thorax dark brown. In some large specimens the head is immaculate above. The hairs are sparse and scarcely more abundant on the thorax than in the variety *occidua.*

FEMALE. Length: 7–8 mm.

Smaller than the female of the typical *rufibarbis.* Surface subopaque and gaster somewhat shining as in the worker. Pubescence longer and denser, hairs more abundant. Head, thorax, and petiole brown; mandibles, cheeks, clypeus, antennae, and legs yellow; mesonotum with three large dark brown blotches, often more or less confluent. Gaster blackish brown. Wings colorless, with dark brown veins and stigma.

MALE. Length 7–8 mm.

Mandibles edentate or indistinctly tridentate. Thorax and gaster stout. Petiole much as in the typical form but with the notch in its superior border often obsolete or narrow.

Surface of body, including the frontal area, opaque; head and thorax coarsely, gaster more finely shagreened; the gaster slightly lustrous.

Hairs extremely sparse, absent on the upper surface of the thorax and gaster. Pubescence grayish, short but rather abundant.

Head and thorax black; gaster dark brown; genital appendages strongly infuscated. Legs yellow. Wings as in the female.

TYPE LOCALITY.— Texas.

Texas: Austin, Fort Davis, New Braunfels, Langtry (Wheeler); Llano (A. W. Morrill); Kerrville, Devil's River (F. C. Pratt).

New Mexico: Las Vegas (T. D. A. Cockerell); Las Valles (Miss Mary Cooper); Mesa Negra, San Ildefonso (E. L. Hewett and Miss Ruth Reynolds); Albuquerque (Wheeler); Alamogordo (G. v. Krockow).

Arizona: Indian Garden, Grand Canyon, Phoenix, Prescott, Tempe, Tucson, Benson Miller Canyon, Huachuca Mountains (Wheeler); Ramsey Canyon, Huachuca Mountains (W. M. Mann).

California: Needles (Wheeler).

Colorado: Canyon City (P. J. Schmitt); Salida (Wheeler).

Utah: Lehi (W. A. Hooker).

This variety has been confounded with *F. fusca* var. *neorufibarbis* and var. *gelida* by Emery, Forel, and myself, owing to the somewhat shining surface of the gaster, but a study of living colonies shows that it belongs to *rufibarbis,* for the workers have the characteristic odor of this species, are aggressive, and live in the ground under stones or in nests without craters. They are found only in shady canyons at rather low altitudes in the Southwest, never in the open desert

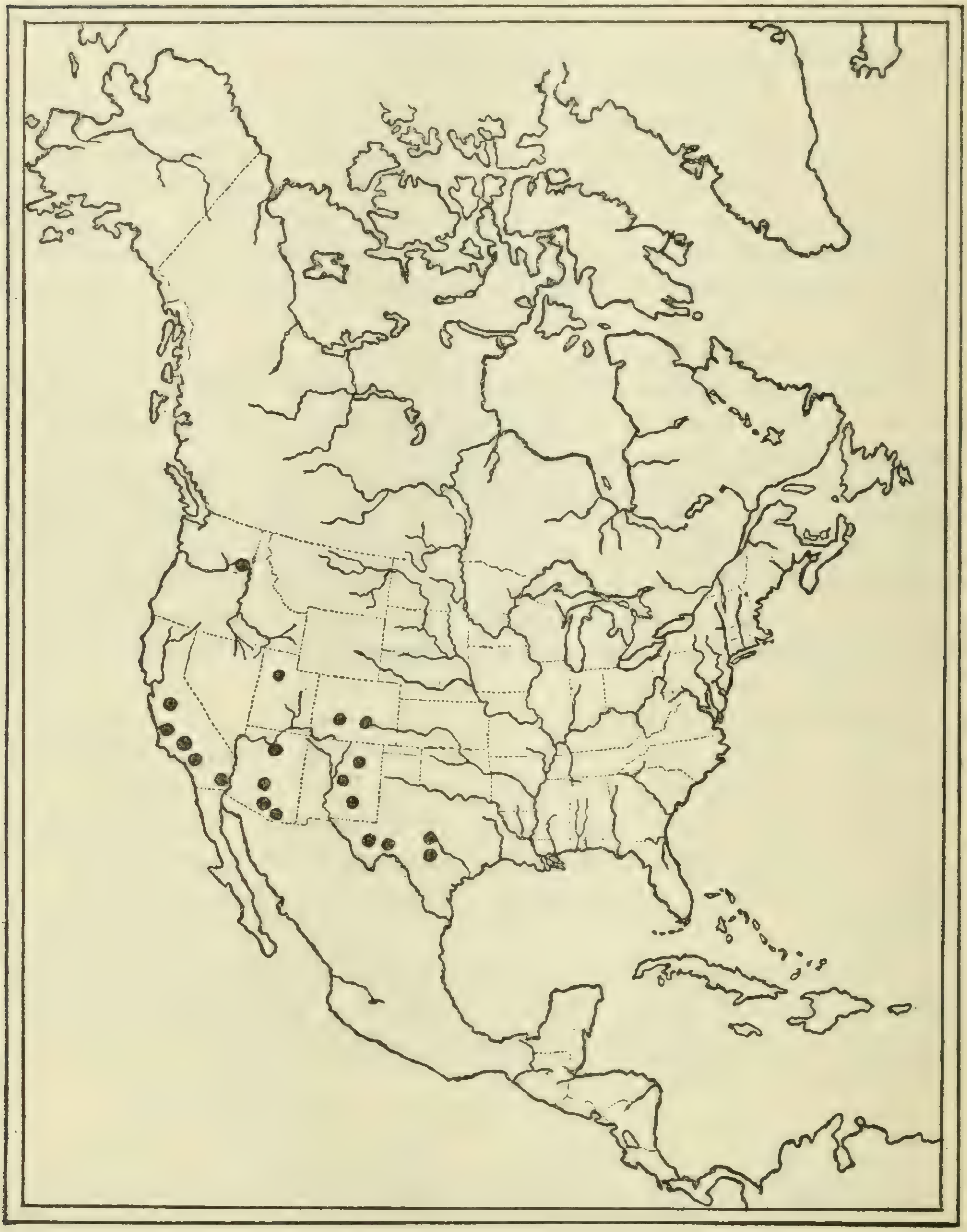

FIG. 7.— Distribution of the Nearctic forms of *Formica rufibarbis*.

country. The colonies which are often rather large, closely resemble those of the variety *occidua* on the Pacific Coast, but the ants, when seen in masses, have a bronzy appearance, as Buckley observed.

103. F. CINEREA CINEREA Mayr.

F. cinerea Mayr, Verh. Zool. bot. ver. Wien, 1853, **2**, p. 280, ☿ ♀; Ibid., 1855, **5**, p. 344, ☿ ♀ ♂; Nylander, Ann. sci. nat. Zool., 1856, ser. 4, **5**, p. 64, ☿ ♀ ♂; Meinert, Natur. abh. Dansk. vid. selsk., 1860, ser. 5, **5**, p. 43, ☿ ♀ ♂; Mayr, Europ. Formicid., 1861, p. 47, 48, ☿ ♀ ♂; Ern. André, Spec. Hymén. Europ. 1882, **2**, pt. 14, p. 181, 186, 189, ☿ ♀ ♂; Lubbock, Ants, bees, wasps, ed. 5, 1882, p. 882, p. 16, etc.; Dalla Torre, Catalog. Hymen. 1893, **7**, p. 193; Ruzsky, Formicar. Imper. Ross., 1905, p. 404.

F. fusca st. *cinerea* Forel, Denkschr. Schweiz. gesell. naturw., 1874, **26**, p. 53, 218, ☿ ♀ ♂; Bruyant, Fourmis France Centr., 1890, p. 56.

F. fusca subsp. *cinerea* Emery, Deutsch. ent. zeitschr., 1909, p. 199, ☿ ♀ ♂.

WORKER. Length 3.5–7 mm.

Very closely related to *F. fusca.* Pro- and mesonotum not very convex, mesoëpinotal constriction rather shallow; epinotum low, with straight base and very sloping declivity, the two surfaces forming a very large and blunt angle with each other. Petiole narrow, with convex anterior and posterior surface and blunt, entire border, which is often produced upward or bluntly pointed in the middle.

Surface of the body opaque, densely shagreened; mandibles somewhat shining, sharply striatopunctate. Frontal area opaque.

Hairs and pubescence white or pale yellow, both very abundant, the hairs short, blunt, and erect or suberect on all parts of the body, except antennal scapes, long on the gula, short and sparser on the legs, and oblique on the flexor surfaces of the tibiae. Pubescence very dense, rather long, uniformly concealing the surface and giving it a silvery appearance.

Dark grayish brown or blackish brown. Mandibles, scapes, legs, and basal halves of funiculi, and in some specimens the middle portions of the femora and tibiae, reddish. In some the reddish tinge also extends to the clypeus, cheeks, petiole, and ventral portion of the thorax.

FEMALE. Length 9–11 mm.

Robust, with large elliptical gaster, superficially resembling the females of *F. fusca* var. *subsericea* and var. *glebaria.* Like the worker in sculpture, pilosity, and color, except that the hairs are longer, more slender and pointed, even on the gaster. The petiole is broad, with flat posterior surface and sharp superior border, often slightly emarginate in the middle. Wings grayish hyaline with pale brown veins and darker stigma.

MALE. Length 8–10 mm.

Body slender, as in the typical *fusca*. Mandibles bidentate or edentate. Head shaped like that of the male of *fusca*. Petiole low and thick, with blunt superior border, which is transverse and feebly and broadly excised.

Body, including the frontal area, opaque, gaster slightly glossy.

Hairs grayish, erect, short, abundant, except on the upper surface of the gaster, oblique on the legs. Pubescence brownish, dense but shorter than in the worker so that the body is less silvery.

Black; gaster dark brown, genitalia and legs yellow; the middle portions of the femora and the genital appendages sometimes infuscated, scapes and mandibles often reddish or yellowish.

Central and Southern Europe and Asia Minor (Caucasus Mountains).

This species nearly always nests in pure sand or sandy soil, preferring river and lake bottoms. It forms huge colonies often extending over many nests, the entrances of which are not surmounted by mounds but only by small, obscure craters. The color of the worker and female is variable, being sometimes as dark as the typical *fusca*, in other colonies more like *rufibarbis*. Specimens of the former coloration were called *fusco-cinerea* by Forel, of the latter *cinereo-rufibarbis*. It is doubtful, however, whether these represent transitions to *fusca* and *rufibarbis*. They may be hybrid forms.

104. F. CINEREA CINEREA var. FUSCO-CINEREA Forel.

F. fusco-cinerea Forel, Denkschr. Schweiz. gesell. naturw., 1874, **26**, p. 55, 57, 58, ☿ ♀ ♂.

F. cinerea var. *fusco-cinerea* Dalla Torre, Catalog. Hymen., 1893, **7**, p. 194.

WORKER AND FEMALE. Intermediate in pilosity and pubescence and also in habits between *F. fusca* and *cinerea*.

MALE. Apparently indistinguishable from the male of the typical *cinerea*.

Zürich and Canton Vaud, Switzerland. Emery does not recognize this form in his revision of the Palaearctic Formicae. It is probably very closely related to the form described below as var. *altipetens* from Colorado.

105. F. CINEREA CINEREA var. IMITANS Ruzsky.

F. cinerea var. *imitans* Ruzsky, Ants envir. Aral Sea (Russian), 1902, p. 10, nota ☿; Zool. jahrb. Syst., 1902, **17**, p. 472; Formicar. Imper. Ross., 1905, p. 405, ☿ ♀.

F. fusca cinerea var. *imitans* Emery, Deutsch. ent. zeitschr., 1909, p. 199.

WORKER. Length 5–6.5 mm.

Pubescence dense, giving the body a silky luster. Erect hairs rather abundant, sparse on the thorax, more numerous on the gaster, long on the gula.

Color pale, resembling that of *F. rufibarbis,* the body being light reddish brown, the gaster dark brown, the legs only slightly darker than the thorax. Head and pronotum above each with a brown spot, or the whole body dark brown above. Small workers darker.

Ssamara and Orenburg in Western Siberia, Kirghis Steppe, Caucasus.

106. F. CINEREA CINEREA var. ARMENIACA Ruzsky.

F. cinerea var. *armeniaca* Ruzsky, Formicar. Imper. Ross., 1905, p. 406, ☿ ♀.

F. fusca cinerea var. *armeniaca* Emery, Deutsch. ent. zeitschr., 1909, p. 199.

WORKER. Length 5–7 mm.

Hairs not as numerous as in the typical *cinerea;* color darker, the head, gaster, and funiculi dark or blackish brown; petiole, scapes, mandibles, and legs brown. Specimens of still darker color and somewhat more abundant pilosity and pubescence occur and form a transition to the typical form.

FEMALE. Length 9–11 mm.

Resembling the typical form except in the gaster, which is hairless and shining.

Caucasus Mountains.

107. F. CINEREA CINEREA var. ALTIPETENS, var. nov.

WORKER. Length 3.5–6 mm.

Shape of thorax like that of the worker *fusca,* with the epinotum more angular and its declivity less sloping in profile, the mesoëpinotal constriction deeper and the pro- and mesonotum more convex. Petiole broad, seen from behind cordate, the border notched in the middle and much sharper than in the typical *cinerea.*

Surface of body a little more shining, being more delicately shagreened. Frontal area opaque. Mandibles subopaque, coarsely striatopunctate.

Hairs less abundant than in the typical *cinerea*, absent on the sides of the head and thorax; only a few long erect hairs on the gula. Legs without erect or oblique hairs on their flexor surfaces. Pubescence dense, but shorter and less silvery than in the typical *cinerea*.

Color of body as in the darker forms of the European type; mandibles, cheeks, anterior border of clypeus, antennae, except the tips of the funiculi, petiole, and legs dark red or brownish.

FEMALE. Length 7 mm.

Closely resembling the worker in sculpture, pilosity, and color. Hairs shorter and less abundant than in the female *cinerea* and as in the worker absent on the sides of the head and thorax. Wings colorless and more transparent than in the typical form, with pale brown veins and darker stigma.

MALE. Length 7–8 mm.

Differing from the male of the typical *cinerea* in the same characters as the worker and female, the body being smoother, less pilose and more delicately pubescent. There are very few erect hairs on the gula and there are none on the legs. Gaster dark brown; genitalia distinctly infuscated. Antennae black, like the head, thorax, and petiole; legs clear yellow. Mandibles very narrow, edentate, with long points, black, with brownish tips. Wings as in the female.

Described from many workers, two males, and a single rather immature female from Florissant, Colorado (8,100 ft.). I have also found this variety on Cheyenne Mountain, near Colorado Springs at about the same elevation. At first sight it would seem to be a hybrid between *F. fusca* var. *argentea* and the next variety, *neocinerea*, but the latter does not occur at Florissant, being peculiar to lower altitudes, and the var. *altipetens* is extremely common in the type locality, where it forms populous colonies which inhabit large earthen mound-nests (2–3 ft. in diameter and 6–10 inches high), overgrown with grass in the alpine meadows. It also nests under stones in the same stations. It is enslaved by *Polyergus breviceps* and the alpine forms of *F. sanguinea*.

108. F. CINEREA CINEREA var. NEOCINEREA Wheeler.

F. cinerea Wheeler, Amer. nat., 1902, **36**, p. 947.
F. cinerea var. *neocinerea* Wheeler, Ants, 1910, p. 571.

WORKER. Length 3–6 mm.

Shape of thorax varying from that of the var. *altipetens* to that of the typical *cinerea*. Petiole more as in the latter form, the border being sharper and broader, but usually entire and sometimes bluntly angular in the middle.

Body but slightly more shining than in *cinerea.*

Pilosity and pubescence more abundant than in the var. *altipetens* but less abundant than in the typical *cinerea,* the erect hairs lacking on the sides of the head, pleurae, and extensor surfaces of the legs as in the former. Pubescence long and dense, but less silvery than in the European form.

Body dark brownish, with the top of the head, the gaster and sometimes the thoracic dorsum darker and more blackish. Antennal scapes scarcely infuscated at their tips.

FEMALE. Length 8–10 mm.

Closely resembling the worker in color, sculpture, and pilosity, but sides of head and thorax with sparse erect hairs as in the female of the typical *cinerea.* Mesonotum with three large fuscous blotches which are confluent behind, the mesopleurae, scutellum, metanotum, and base and sides of epinotum also fuscous. The red color of the anterior part of the head often extending back onto the front. Wings colorless, with pale brown veins and darker stigma.

MALE. Length 7–8 mm.

Closely resembling the male of the var. *altipetens* but the erect hairs on the head and thorax are more abundant and the genital appendages are less infuscated. The antennal scapes, bases of funiculi, and in most specimens also the mandibles are sordid yellow. Wings as in the female.

TYPE LOCALITY.— Illinois: Rockford (Wheeler).

Illinois: New Bedford (G. E. Sanders).

Indiana: Wilders (W. S. Blatchley).

South Dakota: Harding County (S. S. Visher).

Colorado: Breckenridge (P. J. Schmitt); Colorado Springs (Wheeler).

California: San José (H. Heath); Palo Alto, Santa Cruz Mountains (W. M. Mann); Mesa Grande, Russian R. (J. C. Bradley).

In color this variety approaches very closely the redder form of *cinerea* which Forel has called *cinereo-rufibarbis.* Like the variety *altipetens* it nests in meadows and bogs, but its nests, though equally populous, are usually much flatter mounds. This ant is fond of nesting in the natural "hummocks," which are so prominent a feature of the bogs and meadows of Illinois and the neighboring states.

109. F. CINEREA CINEREA var. RUTILANS, var. nov.

WORKER. Length 4–5 mm.

Head large and broad, thorax shaped like that of *fusca,* petiole much compressed anteroposteriorly, with very feebly convex anterior and flat posterior surface, cordate when seen from behind, broad above, narrow below, its edge thin and sharp and narrowly excised in the middle.

Head and thorax opaque; gaster very feebly shining.

Hairs yellow, somewhat less abundant than in the typical *cinerea,* absent on the sides of the head and mesopleurae. Pubescence grayish, dense, but more delicate than is the tyical form, especially on the head and thorax, so that the surface is much more exposed.

Light yellowish red; top of head or at least the ocellar triangle, a large spot on the pronotum, the upper surface of the gaster and the tips of the funiculi brown.

Described from a dozen workers taken at Rockford, Illinois from a single colony occupying a very low mound-nest not unlike those constructed by the var. *neocinerea.* This variety is extremely close to the var. *imitans,* but differs from it in lacking the prominent erect hairs on the sides of the head and thorax and in the slightly less abundant hairs on other parts of the body.

110. F. CINEREA CINEREA var. LEPIDA, var. nov.

WORKER. Length 3.5–6.5 mm.

Very similar in the structure of the thorax and petiole to the typical European *cinerea,* the epinotum being rather low and rounded, especially in smaller workers, and the petiole narrow and blunt.

The sculpture and pilosity are also very similar to the European type, the erect hairs being present on the sides of the head although absent on the pleurae. Legs with small, erect, scattered hairs on their extensor surfaces. The pubescence is dense and glistening white, even more silvery than in the European form, most conspicuous on the gaster though equally dense on the head and thorax.

Color reddish yellow; antennae darker, posterodorsal surface of head blackish brown; gaster brown above, paler than the top of the head and sometimes scarcely darker than the thorax, which may be very feebly infuscated.

Described from numerous specimens taken by Mr. J. C. Bradley at Blue Lake, Humboldt County, California. Except in color, this is the most closely related of all our North American varieties of *cinerea* to the typical form. It also closely resembles the subspecies *pilicornis* in general appearance and color, but may be readily distinguished by the absence of erect hairs on the antennal scapes and eyes and the less abundant pilosity of other parts of the body.

111. F. CINEREA PILICORNIS Emery.

F. fusca var. *cinerea* Mayr, Verh. Zool. bot. ver. Wien, 1886, **36**, p. 427, ☿.
F. pilicornis Emery, Zool. jahrb. Syst., 1893, **7**, p. 664, ☿ ♀ ♂.

WORKER. Length 3–7 mm.

Thorax and petiole very much as in the typical *cinerea;* but in large workers the pro- and mesonotum are very convex and rounded. Petiole rather narrow, thick, with blunt, entire or feebly emarginate superior border.

Body, including the frontal area, opaque; mandibles densely striated and coarsely punctate.

Hairs silvery white, short, pointed, more abundant than in the typical *cinerea,* covering not only the whole body, but also the scapes, legs, and eyes. Pubescence silver gray, very dense and longer than in *cinerea,* uniformly investing the head, thorax, and gaster, much shorter on the legs and scapes.

Brownish red; mandibles darker, tips of funiculi, posterodorsal portion of head and dorsal portion of gaster dark brown, but appearing gray on account of the dense pubescence.

FEMALE. Length 8–10 mm.

Closely resembling the worker in sculpture, pilosity, and color. Three large spots on the mesonotum, the scutellum, metanotum, and sometimes also the pleurae and base of epinotum infuscated. In some specimens the mesothoracic spots become confluent so that the whole dorsal surface of the thorax is fuscous. Wings colorless, with pale yellow veins and darker stigma.

MALE. Length 8–9 mm.

Very similar in color, sculpture, and pilosity to the male of the typical *cinerea,* but the scapes have sparse, erect hairs on their anterior surfaces and the eyes are hairy. The upper surface of the gaster is also sparsely hairy. Body, including mandibles and antennae, black; genitalia and legs yellow; in some specimens the middle portions of the femora are deeply infuscated. Frontal area opaque. Wings as in the female.

TYPE LOCALITY.— California.

California: San Jacinto, Tres Pinos (Th. Pergande); Mount Pinos (F. Grinnell Jr.); Point Loma, San Diego County (P. Leonard and Wheeler); Arroyo Seco at Pasadena, Lakeside (Wheeler); Escondido, San Diego County (J. C. Bradley); Claremont (C. F. Baker); Lake Merced, near San Francisco (F. X. Williams).

This beautiful ant was described as a distinct species by Emery, but it is really only a very pilose subspecies of *cinerea,* peculiar to the

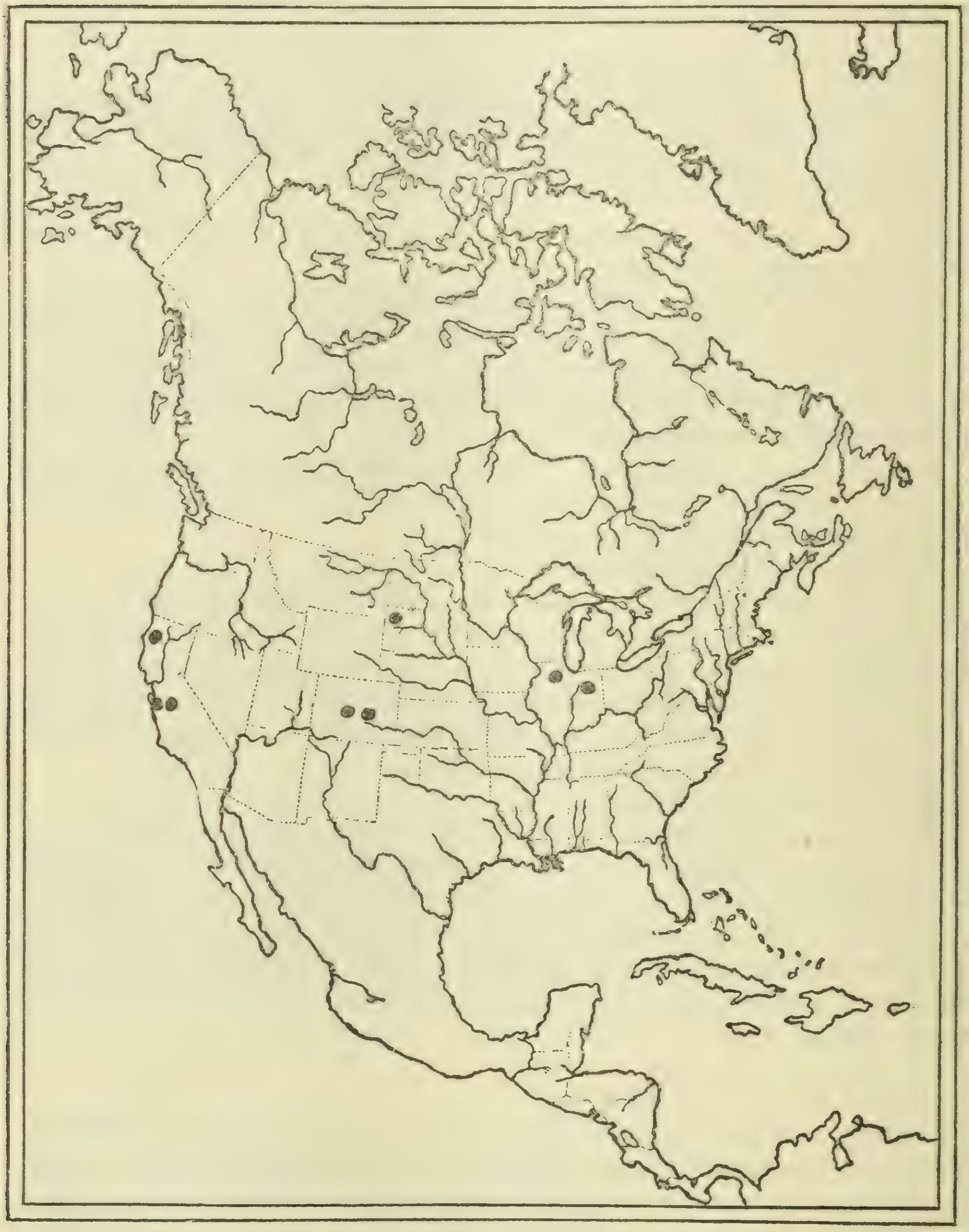

Fig. 8.— Distribution of the Nearctic forms of *Formica cinerea*.

low elevations on the slopes of the Coast Range in California. I have found great numbers of its colonies in the sandy bottom of the Arroyo Seco at Pasadena and in the sandy soil about the lake at Lakeside in El Cajon Valley. In the former locality it was living under large stones, in the latter it formed scattered crater nests, much like those of the typical *cinerea* in sandy portions of the Rhone Valley in Switzerland.

112. F. MONTANA Emery.

F. subpolita var. ? *montana* Emery, Zool. jahrb. Syst., 1893, **7**, p. 663, ☿.
F. subpolita var. *montana* Wheeler, Ants, 1910, p. 571.

Worker. Length 4–4.5 mm.

Closely resembling *F. cinerea* in shape, sculpture, and pilosity. Head longer than broad, narrower in front than behind, with broadly rounded posterior corners and feebly convex sides. Eyes large. Clypeus rather bluntly carinate, its anterior border projecting, entire and broadly rounded. Frontal carinae diverging behind. Antennae moderately long, the scapes slightly enlarged at their tips; joints 2–5 of the funiculus more slender and slightly longer than the penultimate joints. Maxillary palpi rather long. Pro- and mesonotum feebly convex, mesoëpinotal constriction shallow, epinotum with subequal base and declivity, the former straight, forming a blunt obtuse angle with the very sloping declivity. Petiole rather narrow, slightly convex in front, flattened behind, the border not very sharp, seen from behind straight and transverse. Gaster and legs of the usual configuration.

Opaque and very densely shagreened; mandibles striatopunctate, glossy. Frontal area slightly shining.

Hairs pale yellow, abundant, erect, present on the dorsal and gular surface of the head, the thorax, petiole, and gaster; scapes and legs without erect or oblique hairs. Pubescence silvery white, very short, but rather dense, giving the head, thorax, and gaster a pruinose appearance.

Pale reddish brown, posterodorsal portion of head, tips of mandibles and of funiculi somewhat darker.

Type locality.— Nebraska: (Th. Pergande).

Redescribed from one of the cotypes kindly given me by Professor Emery. At first sight this species seems to resemble *F. bradleyi* of the *sanguinea* group, but closer examination discloses many differences. The latter species has a differently shaped head, smaller eyes, a notched clypeus, a shining surface, and much sparser and longer pubescence. I am inclined to believe that *montana* is merely a variety of *cinerea* allied to *rutilans*, but as I have seen only a single specimen, I have merely removed it from *subpolita*, to which it was doubtfully referred by Emery. It certainly has no close relationship to that species.

113. F. SIBYLLA, sp. nov.

WORKER. Length 5–6.5 mm.

Head, excluding the mandibles, somewhat longer than broad, a little narrower in front than behind, with straight sides and slightly convex posterior border. Eyes small. Clypeus very sharply carinate, its anterior border entire, angularly produced in the middle. Maxillary palpi rather long. Frontal carinae slightly diverging posteriorly. Antennae long and slender; scapes very feebly curved at the base, not enlarged distally, funicular joints long and the basal ones slender. Thorax long, pro- and mesonotum only moderately convex, mesoëpinotal constriction shallow, base of epinotum feebly convex, distinctly longer than the sloping, slightly concave declivity. Petiole narrow, thick below, its anterior surface convex below, flattened above, posterior surface flat; border rather sharp, entire or very feebly notched in the middle which is slightly produced upward. Legs rather long and slender.

Opaque; body with a faint bronzy luster in certain lights. Mandibles very finely striated, with scattered, shallow punctures. Clypeus and head, including the frontal area, coarsely and densely, thorax and gaster a little more finely shagreened.

Hairs whitish, long and very sparse on the clypeus, upper surface of head, gula, and fore coxae; shorter and more obtuse on the gaster, except at its tip; almost absent on the thoracic dorsum. Legs without hairs, flexor surfaces of tibiae beset with a sparse row of bristles. Pubescence silvery grayish, very short and dense, covering the head, thorax, petiole, and gaster, even finer and slightly sparser on the legs.

Body black; cheeks, clypeus and legs dark reddish brown; mandibles, antennae, tarsi, and articulations of legs red; tip of terminal funicular joint infuscated.

MALE. Length 9–10.5 mm.

Mandibles broad, with three to four distinct teeth. Head flattened, as broad as long, much narrowed in front, with straight posterior border and cheeks. Eyes narrow, more than twice as long as broad, very convex. Clypeus projecting, distinctly carinate only on its anterior half. Thorax and gaster robust. Petiole low and thick, seen from behind with straight, entire, blunt border. Genitalia considerably withdrawn. Wings long (11 mm.).

Head and thorax, including the mandibles and frontal area, opaque, densely punctate, the pronotum also obscurely reticulate-rugose. Gaster coarsely shagreened, feebly shining.

Hairs and pubescence grayish, both abundant, covering the whole body, the erect hairs on the head, clypeus, gula, mesonotum, scutellum, petiolar border and both the upper and lower surfaces of the gaster being very conspicuous and the pubescence being unusually long, though not sufficiently abundant to conceal completely the ground surface. Eyes hairless.

Black; genitalia and tips of mandibles red; legs yellowish red, with the middle portions of the femora blackened. Wings uniformly and moderately infuscated, with pale brown veins and dark brown stigma.

Described from four workers and two males taken from a single colony by Prof. C. F. Baker in King's Canyon, Ormsby County, Nevada. The specimens were nesting under a log.

At first sight one would not hesitate to regard this ant as *F. fusca* var. *subsericea,* but closer examination shows it to be quite distinct. The eyes of the worker are much smaller than those of *subsericea,* the antennae are more slender, with less curved scapes, it has prominent hairs on the gula and the surface of the body is bronzy and not so black. The male is very different from the male *subsericea* in having smaller eyes, broad, toothed mandibles, a much more pronounced sculpture and very different pilosity and pubescence.

114. F. SUBRUFA Roger.

F. subrufa Roger, Berlin. ent. zeitschr., 1859, **3**, p. 236, ☿; Mayr, Europ. Formicid., 1861, p. 46, ☿; Ern. André, Rev. mag. zool., 1874, ser. 3, **2**, p. 183; Spec. Hymén. Europe, 1882, **2**, pt. 14, p. 181, ☿; Dalla Torre, Catalog. Hymen., 1893, **7**, p. 212; Emery, Zool. jahrb. Syst., 1894, **7**, pl. 22, fig. 20, ☿; Deutsch. ent. zeitschr., 1909, p. 199, ☿ ♀ ♂ ?, fig. 10.

WORKER. Length 5–6.5 mm.

Body slender; head, excluding the mandibles, distinctly longer than broad, narrower in front than behind, with nearly straight posterior and lateral borders and rounded posterior corners. Eyes rather large, convex. Clypeus strongly carinate, its anterior border produced, feebly sinuate or trunate in the middle. Frontal carinae diverging behind. Maxillary palpi long, 6-jointed. Antennae slender, scapes not incrassated towards their tips. Thorax narrow, with moderately rounded pro- and mesonotum and similarly rounded epinotum, separated by a very long, shallow, saddle-shaped mesoëpinotal constriction, so that the thorax is somewhat dumb-bell-shaped both in profile and when seen from above. Petiole narrow, cuneate in profile, thick below, compressed above, but with a blunt border which, seen from behind, is rounded and entire. Legs long and slender. Opaque; surface of body peculiarly shagreened so that it has a silky luster irrespective of the pubescence. Mandibles very finely striated and with small, scattered, indistinct punctures.

Hairs glistening white, short, blunt, erect, abundant, covering the whole body, except the antennae and flexor surfaces of the legs. Pubescence very dilute and delicate, more distinct on the gaster than on the head and thorax.

Ferruginous brown; gaster dark brown; clypeus, cheeks, mandibles, tarsi, and articulations of legs paler, more yellowish red.

Female. Length 9 mm.

Similar to the worker in color and sculpture. Head somewhat broader than the thorax; clypeus feebly carinate, its anterior border more distinctly sinuate than in the worker. Thorax low, flat, with rounded epinotum. Superior border of petiole sharper than in the worker, but much blunter than in the female of *fusca*.

Male (?) Length 7–7.5 mm.; fore wing 5.5 mm.

Black; opaque throughout; tips of mandibles and the genitalia red, the stipes of the latter partly brown. Pubescence on the gaster long, whitish, not very dense; erect hairs very short. Head short and broad. Mandibles opaque; anterior border of clypeus rounded. Thorax depressed. Petiole thick, cuneate, its upper border not excised. Wings nearly colorless, with dark brown veins and stigma.

Iberian Peninsula and Southern France (Eastern Pyrenees).

The worker of this species is easily distinguished by its peculiar sculpture and, as Emery has pointed out, by the singular structure of the thorax in the mesoëpinotal region. I have redescribed the worker from two specimens collected by Prof. G. Strobl at Algeciras in Andalusia. The descriptions of the female and supposed male are taken from Emery.

115. F. SUBPOLITA Mayr.

F. fusca var. *subpolita* Mayr, Verh. Zool. bot. ver. Wien, 1886, **36**, p. 426, ☿ ♀.

F. fusca subsp. *subpolita* Emery, Zool. jahrb. Syst., 1893, **7**, p. 661, ☿ ♀.

F. gagates var. *subpolita* Dalla Torre, Catalog. Hymen., 1893, **7**, p. 199 (*in part*).

F. rufiventris Emery, Zool. jahrb. Syst., 1893, **7**, p. 665, pl. 22, fig. 11, ♂.

F. flammiventris, nom. nov. Wheeler, Psyche, 1912, **19**, p. 90, ♂.

Worker. Length 3–6 mm.

Distinctly dimorphic, the largest workers having the head large, rectangular, as broad as long and only slightly narrower in front than behind, with straight posterior border and feebly convex sides; the small workers having the head much smaller, slightly longer than broad, with straight sides, and posterior borders and more rounded posterior corners. Eyes small; in the largest workers flat, in small workers more convex. Mandibles convex. Clypeus very strongly carinate, its anterior border subangularly produced in the middle. Frontal carinae diverging behind. Antennae stout; scapes rather strongly curved at the base, distinctly incrassated at their tips; funicular joints 2–4 narrower but scarcely longer than the penultimate joints; first funicular joint nearly as long as the second and third together, the second shorter than the third. Maxillary palpi rather short. Thorax short and robust; the pro- and especially the mesonotum very convex in the largest workers, the mesoëpinotal constriction short and deep, the epinotum with the base broadly convex in profile and distinctly shorter than the sloping declivity into which it passes through a rounded angle. In medium sized and small workers, the pro- and mesonotum are only moderately convex and the mesoëpinotal constriction is shallow. Petiole rather high and broad, compressed anteroposteriorly, with convex anterior and flat posterior surface; the border sharp and when seen from behind broadly rounded and entire. Gaster rather large; legs stout.

Surface of body shining, especially the gaster and posterior half of the head, finely shagreened. In the largest workers the mandibles, clypeus, front, cheeks, thorax, and petiole are opaque or subopaque and more coarsely sculptured; the mandibles and clypeus being sharply, densely, and longitudinally striate, the mandibles striatopunctate, the remaining opaque surfaces sharply shagreened. In medium and small workers the anterior portion of the head, including the mandibles, clypeus, and thorax, is distinctly shining and much more delicately shagreened. Frontal area in some specimens opaque, in others smooth and shining, apparently irrespective of the size of the specimen.

Hairs golden yellow, coarse, pointed, erect and very sparse, present on the clypeus, upper surface of the head, gula, pronotum, and gaster. Pubescence short and very sparse, with difficulty perceptible under an ordinary magnification even on the gaster; very fine and dense on the scapes.

Body varying from brownish red to dark chestnut-brown; legs paler and more yellowish; gaster and posterodorsal portion of the head black. Tips of antennal funiculi and sometimes also in large workers the middorsal portion of the pro- and mesonotum infuscated.

Female. Length 8–10 mm.

Resembling the worker, but the whole head opaque, finely and densely punctate behind, with coarsely striatopunctate mandibles and sharply striated clypeus. Frontal area opaque and finely punctate. Thorax subopaque, finely and densely punctate, except the mesonotum and scutellum which are shining and very sparsely punctate. Gaster shining, very delicately shagreened and with minute, scattered piligerous punctures.

Pilosity and pubescence as in the worker.

Color variable. Most individuals have the head, thorax, petiole, and gaster black, the mandibles, legs, and antennae, except the tips of the funiculi, deep red. Others have the venter and base of the first gastric segment and the border of the petiole light red, and still others have the whole gaster and the petiole, except its extreme base, red. Wings grayish hyaline, with brown veins and stigma.

MALE. Length 8–9 mm.

Mandibles edentate or indistinctly bidentate. Head small, broader than long, the posterior border broadly convex, the eyes large. Thorax and gaster robust, the latter flattened. Petiole rather high, somewhat compressed anteroposteriorly, transverse, its border rather sharp, seen from behind straight or feebly excised in the middle, rounded on the sides. Genitalia robust, tips of stipes not extending very far beyond the tips of the other appendages.

Somewhat shining; head and thorax a little more opaque, densely punctate. Frontal area rather smooth and shining in some specimens, in others subopaque.

Hairs and pubescence yellow, the former sparse, erect, distributed much as in the worker, but absent on the upper surface of the gaster. Eyes hairless. Pubescence dense but short and not completely concealing the surface.

Black; gaster and legs bright yellowish red; tips of mandibles reddish. Wings as in the female.

TYPE LOCALITY.— California: San Francisco.

California: Pacific Grove (H. Heath, W. M. Mann, Wheeler); Mount Lowe, summit 6,400 (W. Quayle, Wheeler); Palo Alto, Corte Madera Creek (W. M. Mann); Felton, Santa Cruz Mountains (J. C. Bradley); King's River Canyon (H. Heath); Baldy Peak, San Gabriel Mts. (Brewster, Joos, Crawford); Sierra Nevada, Marine County; Goat Island, San Gregorio.

Washington: Orcus Island (W. M. Mann).

Oregon: Corvallis (T. Kincaid).

British Columbia: Vancouver.

This is not a form of *F. fusca* as Mayr, Emery, and I have been supposing, but a very distinct species peculiar to the Pacific Coast. Its citation from the Eastern States and its allocation with *fusca* and *neogagates* has resulted from a study of the medium and small workers only and a failure to recognize the characters of the largest workers which represent a caste as distinct as the worker major of many species of Camponotus. And, curiously enough, the shape of the head and the small size and flatness of the eyes in this caste remind one vividly of the Camponotus worker major. The male *subpolita* was originally described by Emery as a distinct species (*F. rufiventris*), but Mr. W. M. Mann has taken it on Orcus Island, Washington, flying (though not *in copula*) with females which undoubtedly belong to *subpolita,* and I have taken from colonies of this species at Pacific Grove, Cala., deälated females that have the color of the male, *i. e.* with black head, thorax, and petiole and the gaster of a peculiar yellowish red color.

F. subpolita nests under stones in grassy places, in Washington and Northern California at low elevations but ascends to considerable elevations (6,400 ft.) in the southern part of the latter state. The colonies are rather small and the workers are timid. At Point Joe, near Pacific Grove, I found many nests on the sea-shore and containing great numbers of coccids and pseudoscorpionids.

116. F. SUBPOLITA var. CAMPONOTICEPS, var. nov.

WORKER. Length 3–6.5 mm.

Differing from the typical form in the shape of the head and the color of the largest workers. The head is more distinctly rectangular than in the typical *subpolita,* and, excluding the mandibles, slightly broader than long, not narrower in front than behind, except very close to the insertions of the mandibles, with the cheeks straight behind and convex only anteriorly.

Sculpture of clypeus and head finer than in the typical form. Mandibles more superficially striated and shining. Frontal area smooth and shining in some specimens, opaque in others.

Body and legs yellow or yellowish brown, the posterodorsal portion of the head brown, the gaster blackish brown, the mesonotum with a large dark brown spot, the pronotum with a paler and more indefinite spot. Legs clouded with brown. Mandibles bright red. Smallest workers dark like those of the typical form.

TYPE LOCALITY.— Washington: Wawawai (W. M. Mann).

Washington: Rock Lake (W. M. Mann); Govan (J. A. Hyslop); Almota (A. L. Melander).

The head of the maxima worker of this variety is even more camponotiform than that of the typical *subpolita,* owing to the straight sides and more sudden narrowing at the insertion of the mandibles. I am not certain that the smallest workers described as darker in color belonged to the same colony as the largest specimens. The medium workers are pale in color like the largest.

Subgenus PROFORMICA Ruzsky.

117. F. (P.) neogagates neogagates Emery.

F. fusca var. *gagates* Mayr, Verh. Zool. bot. ver. Wien, 1886, **36**, p. 426, ☿.
F. gagates var. *subpolita* Dalla Torre, Catalog. Hymen., 1893, **7**, p. 199.
F. fusca subpolita var. *neogagates* Emery, Zool. jahrb. Syst., 1893, **7**, p. 661, ☿ ♀ ♂; Wheeler, Bull. Amer. mus. nat. hist., 1904, **20**, p. 306; Occas. papers Bost. soc. nat. hist., 1906, **7**, no. 7, p. 21; Forel, Ann. Soc. ent. Belg., 1904, **48**, p. 153.
F. fusca subpolita Wheeler, Bull. Amer. mus. nat. hist., 1906, **22**, p. 345.

Worker. Length 2.5–5.5 mm.

Head even in the largest workers longer than broad, distinctly narrower in front than behind, with very feebly convex cheeks and posterior border and broadly rounded posterior angles. Eyes rather small, but convex, their long axes decidedly shorter than the distance between their anterior border and the anterior corners of the head. Clypeus sharply carinate, its anterior border angularly produced in the middle. Antennae rather slender, scapes but slightly enlarged towards their tips; first funicular joint as long as the two succeeding joints together; these two joints subequal and each slightly shorter than the penultimate joints. Maxillary palpi moderately long. Frontal carinae not abbreviated, scarcely diverging behind. Thorax slender, pro- and mesonotum moderately convex, mesoëpinotal constriction moderately deep, epinotum with subequal base and declivity, usually rounded, without distinct base and declivity but in some specimens more angular. Petiole narrow, with convex anterior and flat posterior surface and rather blunt border, which is entire and rounded when seen from behind. Gaster small; legs slender.

Surface of body smooth and shining, very finely and superficially shagreened. Mandibles and clypeus very finely and densely, longitudinally striated. In the largest workers the anterior half of the head and the thorax may be subopaque. Frontal area smooth and usually shining.

Hairs white, delicate, erect, rather blunt, abundant and moderately long, present on the upper surface of the head, the gula, whole upper surface of thorax, petiole, and gaster. Pubescence white, very sparse but long and distinct on the gaster and legs, and often also on the head and thorax; very fine and dense on the antennal scapes.

Black or very dark brown, the thorax and petiole often more or less piceous or reddish brown, with paler sutures; the head and gaster usually with bronzy reflections; mandibles, antennae, legs, and sometimes also the cheeks and clypeus red; femora sometimes infuscated in the middle.

Female. Length 6–8 mm.

Clypeus not projecting but with its anterior border truncated or even slightly sinuate in the middle. Resembling the worker in sculpture and pilosity, the hairs, however, longer and more pointed, especially on the gaster. Head, thorax, petiole, and gaster black, or more rarely dark brown; mandibles, clypeus, cheeks, antennae, and legs deep red. In some specimens the antennae and legs are more yellowish and in some the thorax is rich chestnut-brown, clouded with black. Mesonotum and scutellum usually smoother and more shining than the pronotum, pleurae, and epinotum. Wings colorless, with pale brown veins and scarcely darker stigma.

MALE. Length 6–7.5 mm.

Mandibles rather broad, edentate. Head broader than long, very broadly rounded behind, strongly narrowed in front, with large eyes and prominent ocelli. Clypeus indistinctly carinate, with rounded anterior border. Thorax and gaster slender. Petiole thick and broad, its upper border very blunt, broadly rounded and entire when seen from behind. Genitalia slender; stipes narrow and rather pointed, extending a considerable distance beyond the volsellae and sagittae.

Body subopaque; gaster somewhat shining; head finely, thorax more coarsely punctate; frontal area opaque.

Hairs gray, erect, abundant on the upper surface of the head, gula, thoracic dorsum, and mesopleurae, slightly less abundant on the petiole and gaster. Pubescence gray, long, moderately dense on the body, very fine and denser on the appendages.

Black; antennae and gaster dark brown; genitalia and legs yellow; middle portions of femora strongly, genital appendages more feebly infuscated. Wings colorless as in the female, with brown veins and stigma.

TYPE LOCALITY.— Pennsylvania; Beatty, (P. J. Schmitt).

New Jersey: Paterson (Wheeler).

New York: Kiamesha (C. T. Brues); Niagara Falls; Ithaca (Cornell Univ. Coll.).

Connecticut: Kent, Salisbury (W. E. Britton); Norfolk, Colebrook (Wheeler).

Massachusetts: Essex County (G. B. King); Chestnut Hill, Forest Hills (Wheeler).

New Hampshire: White Mountains (W. F. Fiske).

Maryland: (Theo. Pergande).

North Carolina: Gray Beard Mount, Blue Ridge (W. Beutenmüller).

Illinois: Algonquin (W. A. Nason).

S. Dakota: Hill City (Th. Pergande).

Montana: Elkhorn, Nigger Hill (W. M. Mann).

Utah: (Amer. Mus. Nat. Hist. Coll.).

Colorado: Colorado Springs, Colorado City, Manitou, Florissant, Wild Horse, Buena Vista (Wheeler); Boulder (T. D. A. Cockerell).

New Mexico: Las Vegas (E. Tuttle, K. Tipton); Glorieta, Pecos (T. D. A. Cockerell); Albuquerque (Wheeler); Las Valles (Miss Mary Cooper); Alamogordo (G. v. Krockow).

Arizona: Ash Fork, Coconino Forest at the Grand Canyon, Williams (Wheeler); Flagstaff (F. E. Pratt).

Wyoming: Carbon County (Amer. Mus. Nat. Hist. Coll.).

Washington: Almota (A. L. Melander); Pullman (R. W. Doane, W. M. Mann); Wawawai (W. M. Mann).

Ontario: Guelph (Wheeler).

Quebec: Kingsmere (Wheeler).

Nova Scotia: Digby (J. Russell).

British Columbia: Vermillion Pass, 5,000–6,500 ft. (E. Whymper).

Alberta: (E. Whymper).

Emery regarded this ant as a variety of *F. subpolita* but it is certainly quite distinct, though its worker resembles the small workers of the latter species. *F. neogagates,* however, differs in its more abundant, more delicate and paler pilosity and in the proportions of the basal funicular joints of the worker. The male differs greatly from the male *subpolita* in color and in the structure of the genitalia. I had referred the species to Proformica before I noticed that Emery regarded his *F. lasioides,* which is merely a subspecies of *neogagates,* as belonging to this subgenus. In certain respects it is a connecting link between Proformica and the subgenus Formica, the frontal carinae not being abbreviated as in the Old World species of the former group.

F. neogagates is a very timid ant which nests in small colonies under stones in open, often very dry and stony country. In the Rocky Mts. its colonies are abundant at altitudes between 6,000 and 8,000 ft., in the Eastern States it is much rarer and more sporadic and, though preferring the hills of the Appalachian system, may descend almost to sea level. It is, however, properly a subboreal species and even in the latitude of New York is rarely found at low elevations. Like the forms of *F. fusca* it is readily enslaved in all parts of its range by the various subspecies of *F. sanguinea.*

118. F. (P.) NEOGAGATES NEOGAGATES var. MORBIDA, var. nov.

WORKER. Length 3–4 mm.

Differing from the typical *neogagates* in the smaller average size and in color. The body is reddish or brownish yellow, the legs and antennae often paler yellow, the head and mandibles, especially the posterodorsal portion of the former, somewhat darker red, the gaster pale brownish above and posteriorly. Palpi brown. Hairs and pubescence yellowish, the former slightly coarser than in the typical form.

FEMALE (DEÄLATED). Length 6 mm.

Sculpture, pilosity, and pubescence as in the worker; mesonotum and scutellum smooth and shining.

Body brownish yellow; gaster with a short, indistinct, transverse, reddish brown band on each segment; head with a band of the same color between the eyes. Mandibles reddish brown, sutures of thorax fuscous or blackish. Legs and antennae, including the funiculi, concolorous with the body.

Described from nine workers and a single female taken by Rev. P. J. Schmitt, O. S. B., at Lenox, Iowa.

119. F. (P.) NEOGAGATES NEOGAGATES var. VINCULANS, var. nov.

WORKER. Length 2.5–4.5 mm.

In size and pilosity like the preceding variety, in color intermediate between it and the typical *neogagates*. Head, thorax, petiole, legs, and antennae yellowish red, gaster dark brown or blackish, posterodorsal portion of head castaneous, in some specimens nearly as dark as the gaster.

FEMALE. Length 6 mm.

Color and pilosity much as in the worker, but the red of the body deeper and the mesonotum with three very large and nearly confluent, castaneous blotches. Base of first gastric segment red. Wings colorless, with resin-yellow veins and stigma.

Described from five workers taken by myself at Rockford, Illinois, and a single female taken by Mr. W. A. Nason at Algonquin in the same state.

120. F. (P.) NEOGAGATES LASIOIDES Emery.

F. lasioides Emery, Zool. jahrb. Syst., 1893, **7**, p. 664, ☿ ; Wheeler, Ants, 1910, p. 571.

F. (*Proformica*) *lasioides* Emery, Zool. jahrb. Suppl., 1912, **15**, p. 100 *nota*.

WORKER. Length 3.5–4.5 mm.

Differing from the typical *neogagates* in its somewhat smaller size, in color, pilosity, and the somewhat shorter legs and antennae.

Body shining, very finely reticulate-striolate or shagreened and very sparsely and finely punctate. Mandibles finely striate.

Pubescence scarcely visible, extremely short and sparse; hairs erect, rather abundant, white and delicate; gula with long hairs. Legs with somewhat oblique, sparse pubescence. Anterior surface of antennal scapes covered with numerous short, erect hairs.

Yellowish brown; antennae and legs paler; gaster and posterodorsal portion of head darker brown.

TYPE LOCALITY.— South Dakota: Hill City (Th. Pergande).

Colorado: Manitou (Wheeler).

Massachusetts: Wellesley (A. P. Morse); Amherst (Amherst College Coll.).

Professor Emery has very generously given me one of the three cotypes of this ant, which he described in 1893 as a distinct species. This specimen agrees well with the material from Massachusetts except that the latter is somewhat darker in color. As Professor Emery has also sent me cotypes of his *lasioides* var. *picea* (*vetula*) and his *neogagates*, I am able, with the aid of the large amount of material in my collection, to form a definite opinion on the status of these various forms and their relations to one another. Both the typical *lasioides* and its var. *picea* were based on small workers and this evidently led Emery to regard them as representatives of a distinct species. Long series of specimens, however, collected from many localities, show that they differ from the typical *neogagates* and its var. *morbida* merely in having erect hairs on the antennal scapes. The differences mentioned by Emery in the length of the scapes and tibiae are, in my opinion, slight and inconstant.

121. F. (P.) NEOGAGATES LASIOIDES var. VETULA Wheeler.

F. lasioides var. *picea* Emery, Zool. jahrb. Syst., 1895, **8**, p. 335, ☿; Wheeler, Ants, 1910, p. 571.

F. fusca subpolita var. *picea* Wheeler, Occas. papers Bost. soc. nat. hist., 1906, **7**, no. 7, p. 21.

F. lasioides var. *vetula*, nom. nov. Wheeler, Psyche, 1912, **19**, p. 90.

WORKER. Length 2.5–5.5 mm.

Resembling the worker of the typical *neogagates* in size, color (even to the variations of color), sculpture and pilosity, and differing only in having delicate, short, erect, white hairs on the anterior surface of the antennal scapes as in the typical *lasioides*.

FEMALE. Length 6–8 mm.

Differing from the female of the typical *neogagates* only in having short hairs, like those of the worker, on the anterior surfaces of the scapes.

MALE. Length 6–7.5 mm.

Indistinguishable from the male of the typical *neogagates*. Antennal scapes without erect hairs.

TYPE LOCALITY.— British Columbia: Yale, (Dieck).

Ontario: Ottawa (Centr. Exper. Farms. Coll.).

Nova Scotia: Digby (J. Russell).

South Dakota: Hill City (Th. Pergande); Harding County (S. S. Visher).

Montana: Helena, Elkhorn (W. M. Mann).

Colorado: Boulder (P. J. Schmitt, S. Rohwer); Minnehaha Falls, Pike's Peak, 8,400 ft. (T. D. A. Cockerell); Woodland Park, 8,000 ft., Ute Pass, Manitou, Colorado Springs, Florissant, 8,100 ft. (Wheeler).

New Mexico: Beulah, 8,000 ft. (T. D. A. Cockerell); Gallinas Canyon (Miss Mary Cooper); Cloudcroft (H. Skinner).

Nevada: Pyramid Lake (W. M. Mann).

Washington: Pullman (W. M. Mann); Olympia, Friday Harbor (T. Kincaid).

Idaho: Moscow Mt. (J. M. Aldrich).

California: Giant Forest, Sequoia National Park, 6,000–7,000 ft. (J. C. Bradley); Pacific Grove (H. Heath, W. M. Mann, Wheeler).

Illinois: Rockford (Wheeler).

Wisconsin: Milwaukee (C. E. Brown).

Michigan: Porcupine Mts. (O. McCreary).

Connecticut: Colebrook, Norfolk (Wheeler); Stafford (W. E. Britton).

Massachusetts: Blue Hills (Wheeler); Wellesley (A. P. Morse); Essex County (G. B. King); Woods Hole (Miss A. M. Fielde).

Vermont: Lyndon (A. L. Melander).

This variety, as the preceding list shows, has much the same distribution as the typical *neogagates* and is often found in the same localities. It forms colonies of the same size and also nests under stones in open places.

122. F. (P.) LIMATA, sp. nov.

WORKER. Length 3.5–5 mm.

Closely related to *F.* (*P.*) *neogagates*. Head, excluding the mandibles, longer than broad, narrower in front than behind, with convex posterior and straight lateral borders. Eyes large, their long diameter nearly equal to the distance between their anterior border and the anterior border of the head. Clypeus strongly carinate, its anterior border entire and projecting, feebly angular or rounded. Antennae rather slender, scapes scarcely thickened at their tips; first funicular joint a little shorter than joints 2 and 3 together, which are subequal; joints 2–4 more slender but not longer than the penultimate joints. Thorax rather slender, pro- and mesonotum moderately convex, mesoëpinotal constriction moderately deep; epinotum in profile with subequal base and declivity, both rather straight in profile and forming a rounded but distinct obtuse angle with each other. Petiole narrow, not very thick at the base, its anterior surface somewhat convex, the posterior more flattened, the border rather sharp, seen from behind rounded and entire or feebly notched in the middle. Gaster small. Legs rather slender.

Mandibles and clypeus very finely longitudinally striated, the former subopaque, the latter and the remainder of the body very shining, the head, thorax, and gaster distinctly smoother than in *neogagates*, the shagreening of the surface being extremely delicate.

Hairs and pubescence yellowish, very sparse, the former erect and pointed, present on the clypeus, front, vertex and gula, fore coxae and gaster, absent on the thorax and petiole. The pubescence on the gaster is distinct but very sparse, much shorter and denser on the epinotum, legs, and scapes; scarcely perceptible on the head.

Thorax, petiole, legs, and antennae brownish yellow; head, tips of antennal funiculi and gaster dark brown; anterior half of head, including clypeus and mandibles paler and more reddish. Pro- and mesonotum often clouded with fuscous above.

Type locality.— Colorado: Florissant, 8,100 ft. (Wheeler).

Colorado: Ute, Cheyenne Canyon near Colorado Springs (Wheeler).

New Mexico: Las Vegas (Wheeler).

This species is readily distinguished from *neogagates* and small workers of *subpolita* by its much larger eyes, smoother and more shining surface, the sparseness of the hairs on the head and gaster, and their absence on the thorax. The last character, however, is not invariable, for one series of specimens from Cheyenne Canyon has a very few hairs on the mesonotum and posterior border of the epinotum. The specimens from Las Vegas are darker and colored more like *neogagates*. *F. limata* forms small colonies which nest under stones or in crater-nests on dry, stony slopes fully exposed to the sun.

123. F. (P.) NASUTA Nylander.

F. nasuta Nylander, Ann. sci. nat. Zool., 1856, ser. 4, **5**, p. 66, ☿; Ern. André, Spec. Hymén. Europe, 1882, **2**, pt. 14, p. 177, ☿; Forel, Ann. Soc. ent. Belg., 1886, **30**, p. 205, ☿; Nasonow, Arb. Lab. zool. Univ. Moskau, 1889, **4**, p. 61; Dalla Torre, Catalog. Hymen., 1893, **7**, p. 201.

F. aerea Roger, Berlin ent. zeitschr., 1859, **3**, p. 237, ☿; Ibid., 1861, **5**, p. 164.

Myrmecocystus nasutus Emery & Forel, Mitth. Schweiz. ent. gesell., 1879, **5**, p. 449; Emery, Ann. Mus. civ. Genova, 1880, **15**, p. 389 *nota* ☿.

F. (*Proformica*) *nasuta* Ruzsky, Horae Soc. ent. Ross., 1903, **36**, p. 304, ☿; Formicar. Imper. Ross., 1905, p. 421, ☿ ♀ ♂; Emery, Deutsch. ent. zeitschr., 1909, p. 200, ☿ ♀ ♂; Zool. jahrb. Suppl., 1912, **15**, p. 100.

Worker. Length 3–5.8 mm.

Head of small worker elongate, rounded behind, of large worker broader and more rectangular. Clypeus indistinctly carinate. Frontal carinae short and subparallel, frontal furrow distinct only in large individuals. Thorax long and low; its dorsal contour in profile angularly impressed in the mesoëpinotal region only in the larger workers; seen from above the pronotum is broader than the epinotum, twice as broad as the mesonotum and spherically convex. Petiole higher than broad, its border more rounded in small individuals, angularly excised in large specimens.

Body very shining; front and clypeus finely longitudinally striated, subopaque.

Erect hairs long and sparse; pubescence very sparse, in some specimens more abundant on the gaster and somewhat concealing the shining surface.

Brown; larger specimens piceous, often with faint metallic luster; mandibles, antennae, and legs paler.

Female. Length, 7 mm., without the gaster 4.3 mm.

Head rather square, scarcely longer than broad, the scapes extending but little beyond the posterior corners of the head. Thorax narrower than the head, with parallel sides and flat dorsal surface. Petiole high, with sharp dorsal border, deeply and angularly excised in the middle.

Sculpture as in the larger workers; pubescence denser, gray; color somewhat darker.

Male. Length 6–7.5 mm., of fore wings 5.5–6.2 mm.

Head rounded, somewhat longer than broad, the eye about $\frac{2}{5}$ the length of the lateral border. Petiole thick, its border obtuse, and seen from behind excavated in the middle. Gaster, excluding the genitalia, as long as the thorax, with distinct intersegmental constrictions; subgenital plate obtusely trilobed. Stipes slender projecting far beyond the volsellae and sagittae. Wings with a well-developed discoidal cell.

Surface of body smooth and very shining.

Hairs brown, long, very dense on the head, thorax, and venter; sparser on the dorsal surface of the gaster; legs with long, reddish, suberect hairs.

Black; funiculi, legs, and borders of gastric segments yellowish brown; genitalia brownish yellow. Wings faintly tinged with brown.

Type locality.— Southern France.

Iberian Peninsula, Balkan Peninsula, Southern Russia, Aralocaspian Plain.

Emery calls attention to the fact that the range of this species is discontinuous, consisting of an eastern and a western region, widely separated from each other, and he admits his inability to detect any differences between eastern and western workers. He has seen male specimens from France and Spain and these agree closely with Ruzsky's description of oriental specimens. Forel, however, is inclined to believe that the eastern and western forms differ somewhat and he therefore refers at least some of the former to the variety *striaticeps*.

124. F. (P.) NASUTA var. STRIATICEPS Forel.

F. (*P.*) *nasuta* var. *striaticeps* Forel, Bull. Soc. Vaud. sci. nat., 1911, ser. 5, **47**, p. 352, ☿.

Worker. Length 3–7 mm.

Differing from the typical form of *nasuta* from Southern France in having the head finely striated almost as far back as the occiput, and more abundantly punctate. The pubescence is also more distinct and more abundant, but nevertheless variable. Color paler and more brownish. The typical form of the species is a little smaller, almost devoid of pubescence and has only the clypeus and a part of the front striated; this sculpture, too, is usually feebler and the punctuation sparser.

Type locality.— Vicinity of Salonica.

Bulgaria, Tiflis, and the Caucasus.

Should further study prove that all the smoother eastern forms of *nasuta* belong to *nasuta*, the name *striaticeps* would have to be replaced by *aerea*, since Roger's types of this form came from Greece.

Forel states that the nests of *striaticeps* are feebly populated and that they are excavated in the earth of rather dry fields or in rocky places. The workers, which leave the nests at infrequent intervals, often return with the gaster much distended with liquid food after the manner of the species of Prenolepis.

125. F. (P.) KORBI Emery.

F. (*P.*) *korbi* Emery, Deutsch. ent. zeitschr., 1909, p. 202, ☿ ♀; Forel, Bull. Soc. Vaud. sci. nat., 1911, ser. 5, **47**, p. 353.

WORKER. Length 2.5–6.5 mm.

Structure and color of the body almost exactly the same as in *nasuta*.

Whole body, even in small specimens, opaque, owing to its fine sculpture, with a slight bronzy or steel-blue luster, most distinct on the gaster.

Pubescence dense, whitish and appressed. Hairs suberect, long, sparse, and obtuse.

FEMALE. Also very similar to *F. nasuta*. Pubescence as in the worker; color black, appendages paler.

Sultan-Dagh in Anatolia (M. Korb).

Forel believes that this form may eventually prove to be only a subspecies of *nasuta*.

126. F. (P.) MONGOLICA Emery.

F. nasuta subsp. *mongolica* Emery, Zichy III Asiat. forschungsreise, 1901, p. 159, ☿.

F. (*Proformica*) *mongolica* Emery, Deutsch. ent. zeitschr., 1909, p. 202.

WORKER. Length 2–4.2 mm.

Resembling the small worker of *nasuta* in color and sculpture, but the head is proportionally broader, the antennae shorter and thicker, the thorax much stouter, and the pronotum narrower in proportion to the posterior portion of the thorax. Pubescence sparse as in *nasuta*.

Chara-Gol in Mongolia.

Emery saw only a few specimens of this species and it is therefore doubtful whether larger ones occur.

127. F. (P.) ABERRANS Mayr.

F. aberrans Mayr, Fedtschenko's Turkestan. Formicid. 1877, p. 7, ☿; Tijdschr. ent., 1880, **22**, p. 27; Ern André, Spec. Hymén. Europe, 1882, **2**, pt. 14, p. 178, ☿; Dalla Torre, Catalog. Hymen., 1893, **7**, p. 192.

F. (*Proformica*) *aberrans* Ruzsky, Formicar. Imper. Ross., 1905, p. 421; Emery, Deutsch. ent. zeitschr., 1909, p. 203.

WORKER. Length 5.5 mm.

Head longer than broad, much rounded behind. Clypeus carinate, its anterior border faintly emarginate in the middle. Frontal carinae subparallel and nearly straight. Petiole thick, with blunt, rounded superior border.

Slightly lustrous; head longitudinally, thorax in part transversely, finely, and sharply striolate; mandibles striatopunctate, gaster delicately transversely striolate.

Hairs suberect, moderately abundant; scapes and tibiae with short, whitish, suberect hairs; pubescence very sparse.

Black; mandibles, antennae, and legs brown.

Turkestan.

128. F. (P.) ABERRANS var. NITIDIOR Forel.

F. (*P.*) *aberrans* var. *nitidior* Forel, Ann. Mus. St. Petersbourg, 1904, **8**, p. 383, ☿ ; Ruzsky, Formicar. Imper. Ross., 1905, p. 421, ☿ ; Emery, Deutsch. ent. zeitschr., 1909, p. 203, ☿ .

WORKER. More shining than the typical *aberrans* and no more sharply sculptured than *F. gagates*.

Turkestan.

129. F. (P.) OCULATISSIMA Forel.

F. oculatissima Forel, Ann. Soc. ent. Belg., 1886, **30**, C. R. p. 161, ♂; Dalla Torre, Catalog. Hymen., 1893, **7**, p. 203.

F. (*Proformica*) *oculatissima* Emery, Deutsch. ent. zeitschr., 1909, p. 203, ♂.

MALE. Length 7 mm.; anterior wings 7.3 mm.

Body slender; head small, with enormous eyes and ocelli. Mandibles edentate. Clypeus ecarinate; clypeal fovea distinctly separated from the antennal fovea. Thorax rather depressed, epinotum more sloping than in the other species of Proformica. Petiolar scale as thick as high, feebly emarginate above. Subgenital plate emarginate on each side, with rounded median lobe. Wings with a large discoidal cell.

Body shining.

Hairs suberect, rather abundant on the head, ventral surface of the body and genitalia, sparse on the thoracic dorsum and upper surface of the gaster, absent on the scape; tibiae with a few suberect hairs.

Black; antennae, mandibles, and coxae yellowish brown, remainder of legs yellow. Wings nearly colorless, with pale veins and brown stigma.

Greece.

130. F. (P.) KRAUSSI Forel.

F. kraussii Forel, Mitth. Schweiz. ent. gesell., 1895, **9**, ☿; Emery, Bull. Soc. ent. France, 1899, p. 18; Forel, Ann. Soc. ent. Belg., 1902, **46**, p. 155, ♂.
F. (*Proformica*) *kraussi* Emery, Deutsch. ent. zeitschr., 1909, p. 204, ☿ ♂.

WORKER. Length 3.2 mm.

Habitus more like that of Lasius than Formica. Head oval, narrower anteriorly; frontal carinae very short. Mandibles 5-toothed, with oblique border. Clypeus very bluntly carinate. Pronotum, seen from above, rounded, spherical, twice as broad as the mesonotum. Petiolar scale truncate, with rounded dorsal border.

Very shining, head less so, very finely rugulose; mandibles finely striated.

Surface scarcely pubescent, with numerous short, erect, obtuse, slightly clavate hairs. Legs with short, sparse pubescence.

Dark brown, gaster piceous with feeble bronze reflections.

MALE. Length 3.5 mm.

Body short. Eyes not especially large; mandibles narrow and pointed. Thorax high; petiole with bluntly rounded node. Gaster short. Volsellae of genitalia very short and slender. Anterior wings with large stigma and without a closed discoidal cell.

Body scarcely pubescent; suberect hairs as in the worker; tibiae with a few oblique bristles.

Piceous, head and tip of gaster almost black; appendages more or less reddish, femora darker.

Known only from Southern Algeria.

131. F. (P.) EMMAE Forel.

F. (*P.*) *emmae* Forel, Bull. Soc. Vaud. sci. nat., 1909, ser. 5, **45**, p. 381, ☿; Emery, Zool. jahrb. Suppl., 1912, **15**, p. 103, ☿.

WORKER. Length 3–5.5 mm.

Mandibles 5-toothed, the apical tooth long; head as broad as long, with the posterior angles rounded and the posterior border scarcely convex. Fourth joint of maxillary palpi curved, as long as the two last joints together; third nearly as long as the fourth. Ocelli distinct. Eyes large, rather convex, situated at the posterior third of the head. Clypeus feebly carinate, the carina interrupted at its anterior third by a small transverse impression. Anterior border of clypeus straight, not projecting. Antennal scapes surpassing the posterior border of the head slightly even in the largest workers. First funicular joint $1\frac{1}{3}$ times as long as the second, all the other joints at least twice as long as broad. Pronotum large; mesonotum very distinctly concave and saddle-shaped in profile, with an obtuse projection in front resembling the pommel of a saddle and a feeble posterior thickening formed mainly by the two projecting spiracles. Epinotum rather broad, with strongly convex basal surface, rising behind the mesonotum and a little longer than the declivity into which it passes insensibly. Petiole vertical, thick, much thicker than in *F. nasuta,* as thick above as at the base, convex in front, flat behind, with obtuse, rounded border, entire and rather transverse when seen from behind. Legs very slender.

Mandibles striatopunctate, rather shining. Body very shining; gaster nearly smooth, other portions of the body very feebly shagreened, a little more strongly and densely on the front of the head.

Front of clypeus with a row of long hairs; remainder of body glabrous, except for one or two small hairs on the head and towards the tip of the gaster. Mentum with ammochaetae. Tibiae with one, metatarsi with two rows of bristles on their flexor surfaces. Pubescence very short and very sparse on the appendages, almost *nil* on the body.

Black, scarcely tinged with brown. Mandibles, anterior border of head, antennae, legs, palpi, and the very narrow posterior border of each gastric segment brownish red; coxae and femora darker brown.

Biskra, Algeria (A. Forel).

This ant is remarkably like the species of Cataglyphis, especially in the structure of the thorax, petiole, and maxillary palpi and in possessing ammochaetae on the mentum. According to Forel, its colonies are very small and inhabit little nests the entrances of which do not open on craters but are very small round holes in the sand, difficult to discover unless one follows up workers that are returning to the nest.

SUBGENUS NEOFORMICA, subgen. nov.

132. F. (N.) PALLIDEFULVA PALLIDEFULVA Latreille.

F. pallidefulva Latreille, Hist. nat. fourm. 1802, p. 174, ☿; Dalla Torre, Catalog. Hymen., 1893, **7**, p. 203; Emery, Zool. jahrb. Syst., 1893, **7**, p. 656, taf. 22, fig. 16, ☿ ♀ ♂; Ibid., 1895, **8**, p. 335; Wheeler, Bull. Amer. mus. nat. hist., 1904, **20**, p. 369.

WORKER. Length 4.5–6 mm.

Body long and slender. Mandibles 8-toothed. Head, excluding the mandibles, about 1¼ times as long as broad, but little narrower

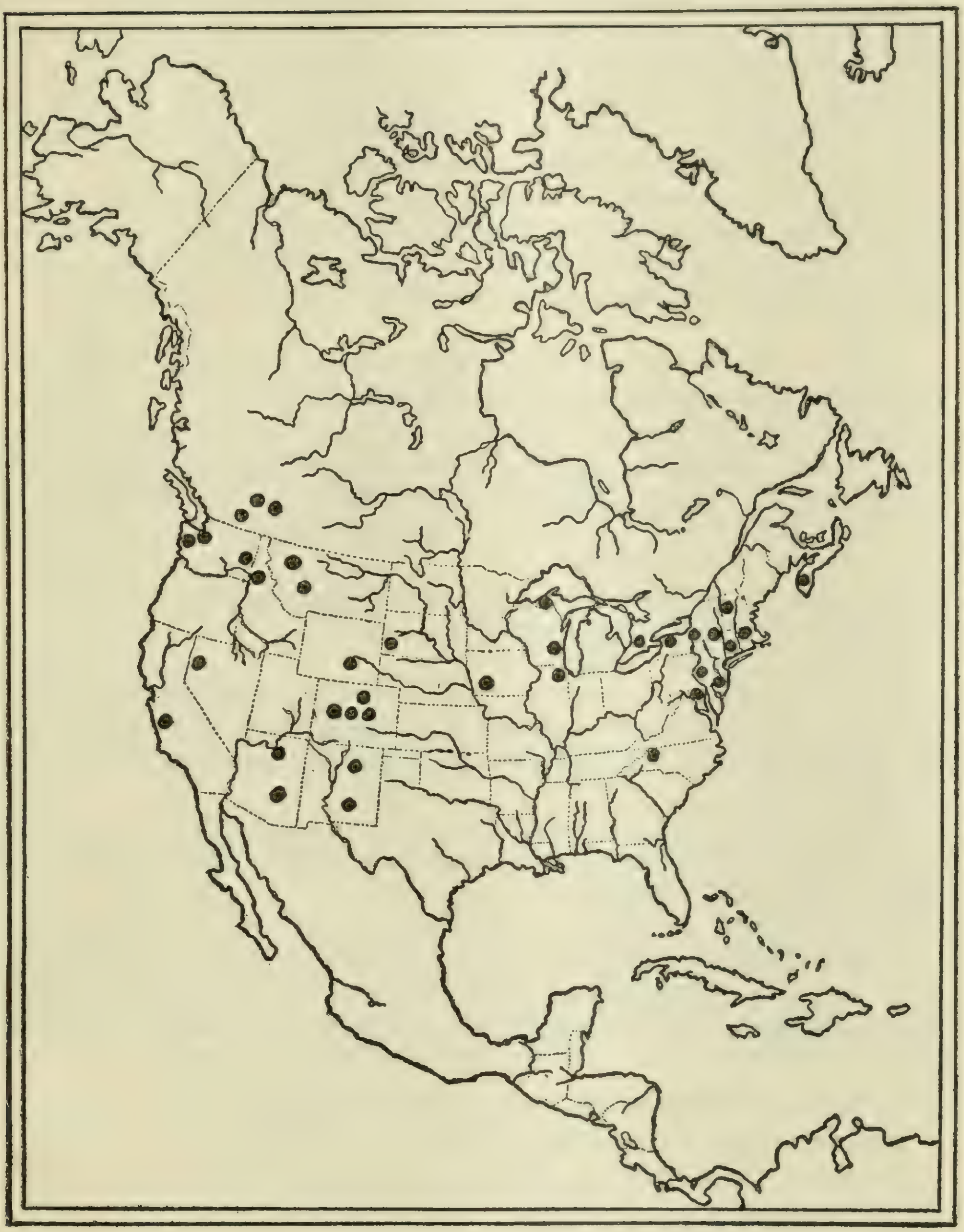

FIG. 9.— Distribution of the subgenus Proformica in North America.

in front than behind, with convex and rounded posterior border and straight sides. Eyes moderately large, convex. Clypeus with broadly rounded and somewhat projecting, entire, anterior border, strongly carinate, the carina angular in profile. Frontal carinae parallel. Maxillary palpi very long, 6-jointed. Antennae very long and slender, the scapes straight at the base, funiculi not thickened towards their tips, the median joints more than 1½ times as long as broad. Thorax, especially the pro- and mesonotum, long, these portions not very convex, evenly rounded; mesoëpinotal constriction not deep; epinotum low and evenly rounded, without distinct base and declivity. Petiole narrow, rather thick, its anterior and posterior surfaces convex, its margin blunt, rounded and entire when seen from behind or only slightly impressed in the middle. Gaster elongate elliptical; legs long.

Whole body shining and very finely and superficially shagreened; mandibles finely striated and coarsely punctate.

Hairs yellowish, sparse, confined to the gaster and the upper surface of the head; absent on the gula, thorax, and petiole; blunt and coarse on the gaster. Legs with only the graduated series of bristles on the flexor surfaces of the tibiae. Pubescence fine and rather dilute, visible on the gaster and legs.

Pale yellow; mandibles reddish; gaster often with a brownish tint; sometimes the whole head is reddish.

FEMALE. Length 7–8 mm.

Rather stout; head nearly as broad as long, subrectangular, a little narrower than the thorax. Border of clypeus more flattened than in the worker, petiole more compressed anteroposteriorly, with flat posterior surface.

Sculpture and pilosity as in the worker, but the body, especially the head and mesonotum, more reddish. Wings colorless, with pale brown veins and stigma.

MALE. (After Emery).

Characterized by its pale color. The whole body is yellow, the head and the gaster behind somewhat darker. Vertex and tips of funiculi brown; only the eyes black. Head much smaller than in the other subspecies. The eyes too are somewhat smaller.

District of Columbia: Washington (Th. Pergande).

Kansas: Osage City (A. C. Burrill).

Georgia: Tallulah Falls, Marietta, Ducker, Clayton, Clarkesville (J. C. Bradley).

New Jersey: Fort Lee (W. Beutenmüller).

New York: Bronxville (Wheeler).

I have not seen the male of this the typical form of the species but the male of the var. *succinea* described below is probably very similar.

Both are southern forms, the true *pallidefulva* being very rare as far north as New Jersey and New York. The species is very constant morphologically although it varies greatly in color, and pilosity. The males of all the forms I have seen are very slender, have very large eyes and resemble the males of the subgenus Proformica in having the stipes of the genitalia projecting considerably beyond the other appendages. What Mayr described as the female of *pallidefulva* from New Jersey, I believe with Emery to be the female of *F. difficilis.*

133. F. (N.) PALLIDEFULVA PALLIDEFULVA var. SUCCINEA Wheeler.

F. pallidefulva var. *succinea* Wheeler, Bull. Amer. mus. nat. hist., 1904, **20**, p. 369, ☿.

WORKER. 4.5–6 mm.

Differing from the worker of the typical form in color, being throughout of a richer, purer, more reddish yellow, and in having the pubescence on the gaster even shorter and more inconspicuous. The whole surface of the body seems to be somewhat smoother than in the typical form and the integument harder.

FEMALE. Length 8–9 mm.

Whole body red, decidedly darker than the worker; mandibles more brownish, legs more yellowish. Wings colorless, with pale brown veins and stigma.

MALE. Length 8–10 mm.

Body long and slender. Head small, with very large eyes, broadly rounded behind. Cheeks very short, straight. Mandibles pointed, edentate, their blades rather broad. Clypeus carinate. Frontal carinae diverging. Maxillary palpi 6-jointed. Antennae very slender. Thorax narrowed in front. Petiole very thick and low, with a very blunt border, which, seen from behind, is transverse and feebly notched in the middle. Gaster long and slender. Stipes of genitalia long and slender, considerably surpassing the other appendages.

Head, thoracic dorsum, and epinotum subopaque; pleurae and gaster shining. Hairs very short, confined to the thoracic dorsum and top of head.

Honey yellow; ocellar triangle black; mesonotum streaked with brownish. Wings colored as in the female.

TYPE LOCALITY.— Texas: Austin (Wheeler).

Texas: Montopolis, Milano, Bee Creek (Wheeler); Victoria (Hunter).

Oklahoma: Ponca City (A. C. Burrill).

I have taken this form in the sandy or pebbly soil of the Texan post-oak woods and among the limestone hills of Travis County. It rarely nests under stones but constructs craters two to four inches in diameter, with a central opening $\frac{1}{2}$–$\frac{3}{4}$ inches in diameter, made of coarse sand or pebbles. These nests resemble those of certain subspecies of *Myrmecocystus melliger* in Colorado, Western Texas, and Mexico. The beautiful yellow males and red females were taken May 26.

134. F. (N.) PALLIDEFULVA SCHAUFUSSI Mayr.

F. schaufussi Mayr, Sitzb. K. akad. wiss. Wien, 1866, **53**, p. 493, fig. 6, ☿; Verh. Zool. bot. ver. Wien., 1870, **20**, p. 951; McCook, Proc. Acad. nat. sci. Phil., 1880, p. 377; Ibid., 1887, p. 29; Dalla Torre, Catalog. Hymen., 1893, **7**, p. 212.

F. pallidefulva subsp. *schaufussi* Emery, Zool. jahrb. Syst., 1893, **7**, p. 654, taf. 22, figs. 17, 18, ☿; Wheeler, Bull. Amer. mus. nat. hist., 1904, **20**, p. 370.

Worker. Length 5–7 mm.

Averaging somewhat larger than the preceding forms of the species, and differing also in having the two terminal joints of the maxillary palpi somewhat shorter and in pilosity and color. The erect hairs are longer and more numerous and are not only present on the gaster and upper surface of the head, but also on the gula, thoracic dorsum, and petiole. The pubescence is much longer and more distinct, especially on the gaster. The color of the head and thorax is more brownish yellow, the gaster more or less infuscated. The mandibles are red.

Female. Length 8–10 mm.

Head, excluding the mandibles, as broad as long, rarely a little longer than broad. Thorax sometimes more slender than in the female of the typical *pallidefulva* and its variety *succinea*. Petiole broad, flattened behind, with entire, broadly rounded border. Sculpture, pilosity, and color as in the worker, hairs very long and abundant on the upper surface of the body, on the gula and petiolar border and especially long on the gaster. Mesonotum usually with three large, elongate brown spots, and the gaster with the bases and posterior borders of the segments a little more brownish than their central portions. In some specimens the whole of the mesonotum and gaster is deep brown. Wings hyaline, sometimes slightly tinged with yellow; veins and stigma pale brown.

Male. Length 8–9 mm.

Mandibles edentate or obscurely bidentate, with rather narrow blades. Head as in the typical form of the species. Petiole low, thick in profile below, with a rather sharp upper border, which is distinctly excised in the middle; both its anterior and posterior surfaces sloping. Gaster more shining than the head and thorax. Hairs rather short, pubescence short, rather dense on the gaster, indistinct elsewhere.

Head black, thorax, antennae, petiole, and gaster dark brown, genitalia scarcely paler than the gaster. Sutures of thorax, legs, and tips of mandibles brownish yellow. Wings as in the female.

Ontario: Toronto (R. J. Crew).

Maine: Ogunquit (H. S. Pratt).

New Hampshire: Durham (C. M. Weed).

Massachusetts: Andover, South Natick, Sherborn (A. P. Morse); Mt. Tom (G. B. King, Geo. Dimmock); Woods Hole, Boston (Wheeler).

Rhode Island: Providence (Davis).

Connecticut: New Haven (W. E. Britton, H. L. Viereck); Salisbury, Stafford (W. E. Britton); Winsted, Norfolk, Colebrook (Wheeler).

New York: Bronxville, Mosholu (Wheeler); West Farms (J. Angus).

New Jersey: Lakehurst, Ramapo Mountains, Weasel Mount, Great Notch (Wheeler); Alpine, Ft. Lee (W. Beutenmüller); Lucaston.

Pennsylvania: White Haven (J. C. Bradley); Chestertown (E. G. Vanatta); Lehigh Gap.

North Carolina: Black Mountains (W. Beutenmüller); Lake Toxaway (Mrs. A. T. Slosson).

Indiana: Pine, Shoals, Hammond, Wyandotte, New Harmony (W. S. Blatchley).

Illinois: Rockford (Wheeler).

Wisconsin: Milwaukee (C. E. Brown).

This is a very common form throughout the Northern States east of the Mississippi. It forms small or moderately large colonies which nest under stones or in obscure crater nests in open, sunny fields and pastures and on grassy hill-slopes. It is an extremely timid ant, usually fleeing with great precipitation when its nest is disturbed, never stopping to defend itself and returning to secure its brood in a furtive and hesitating manner. It lives largely on dead insects and the excreta of aphids. I agree with Emery that the male and female described by Mayr as belonging to this form may be more properly referred to the subsp. *nitidiventris*.

135. F. (N.) PALLIDEFULVA SCHAUFUSSI var. DOLOSA Wheeler.

F. pallidefulva schaufussi var. *meridionalis* Wheeler, Bull. Amer. mus. nat. hist., 1904, **20**, p. 370, ☿.
F. pallidefulva schaufussi var. *dolosa*, nom. nov. Wheeler, Psyche, 1912, **19**, p. 90.

WORKER. Length 5–7 mm.

Resembling the typical *schaufussi* in all respects except that the gaster is scarcely darker than the remainder of the body and the pubescence on the gaster is much longer and denser so that it appears more opaque.

FEMALE. Length 9–10 mm.

Differing from the female of the typical *schaufussi* in the same characters as the worker and also in the coloration of the mesonotum, which is immaculate and of the same brownish yellow tint as the remainder of the body.

TYPE LOCALITY.— Texas: Bull Creek (Wheeler).

Texas: near Austin (Wheeler); Arlington (W. E. Hinds); Edna (J. D. Mitchell).

Arkansas: McNeil (J. D. Mitchell).

Louisiana: Gilliam (F. C. Bishop), Mansfield (W. E. Hinds).

Missouri: Doniphan (P. J. Schmitt).

North Carolina: (P. J. Schmitt).

Georgia: Atlanta, Gainesville, Black Rock Mountain, Rabun County, 3,500 ft. (J. C. Bradley).

This is a distinctly southern variety of *schaufussi*, apparently constant, to judge from the specimens I have seen. I have found it nesting in obsure crater nests in grassy places in the dry canyons of Central Texas. Its habits are essentially like those of the northern *schaufussi*.

136. F. (N.) PALLIDEFULVA SCHAUFUSSI var. INCERTA Emery.

F. pallidefulva schaufussi var. *incerta* Emery, Zool. jahrb. Syst., 1893, **7**, p. 655, ☿ ♀ ♂; Wheeler, Bull. Amer. mus. nat. hist., 1904, **20**, p. 370; Ibid., 1906, **22**, p. 52; Ibid., 1907, **23**, p. 37.

WORKER. Length 4.5–7 mm.

This form differs from the typical *schaufussi* merely in the slightly less abundant pilosity. The hairs on the gula and petiole are few, and may be lacking on one of these regions but very rarely on both. The pubescence is often somewhat shorter and sparser but there seems to be no very constant difference in coloration, although in general the gaster is often fuscous or even blackish and the head and thorax may have a deeper, more brownish or reddish tint.

FEMALE. Length 8–9 mm.

Color, as a rule, darker than in the female *schaufussi*. In addition to the three dark spots on the mesonotum, the gaster and the posterior portion of the head may be dark brown, the former sometimes blackish. Hairs and pubescence sparser and shorter than in the type, gaster smoother and more shining.

MALE. Length 7–9 mm.

Indistinguishable from the male of the typical *schaufussi*.

TYPE LOCALITY.— District of Columbia (Th. Pergande).

Virginia: (Th. Pergande).

New Jersey: Lakehurst, Weasel Mt. (Wheeler); Alpine, Ft. Lee (Wm. Beutenmüller).

New York: New York (C. T. Brues); West Farms (J. Angus); Bronxville, Tuckahoe (Wheeler); Niagara Falls, Arlington, Staten Island (Wheeler).

Pennsylvania: Ashbourne; Lehigh Gap.

Connecticut: Colebrook, Winsted, Norfolk (Wheeler); Bradford (Winkley); Rockville (H. L. Viereck).

Massachusetts: Wellesley, Sherborn (A. P. Morse); Boston (Wheeler); East Northfield (A. C. Burrill).

New Hampshire: Durham (C. M. Weed).

Illinois: Rockford (Wheeler).

Wisconsin: Racine, Milwaukee (C. E. Brown).

Colorado: Colorado Springs, Cheyenne Canyon (Wheeler).

New Mexico: Las Valles (Miss Mary Cooper).

This variety, which lives in the same situations and has the same habits as the typical *schaufussi*, though ranging considerably further west, is, as Emery observed, very unstable or variable both in color and pilosity. Some pale specimens are almost indistinguishable from *schaufussi* while others are smaller, more deeply colored and have so few hairs and such short pubescence that they are equally close to *nitidiventris*. Moreover, such different forms are often present in the same colony.

137. F. (N.) PALLIDEFULVA NITIDIVENTRIS. Emery.

F. schaufussi Mayr, Verh. Zool. bot. ver. Wien, 1886, **36**, p. 427, ♀ ♂.

F. schaufussi subsp. *nitidiventris* Emery, Zool. jahrb. Syst., 1893, **7**, p. 656, taf. 22, figs. 13, 19, ☿ ♀ ♂; Wheeler, Bull. Amer. mus. nat. hist., 1904, **20**, p. 37.

WORKER. Length 4–6 mm.

Differing from the preceding forms of *schaufussi* in its smaller average size, pilosity, pubescence, and usually also in coloration. The hairs, though present on the upper surface of the head and thorax and on the gaster, are lacking on the gula and petiole. The pubescence is extremely short and sparse, so that the gaster is much more shining. The head, thorax, petiole, and appendages are often red or brown and much darker than in *schaufussi* and the gaster is dark brown. Often also the back of the head is darker than the thorax.

FEMALE. Length 6.5–8 mm.

Smaller than the female of the preceding forms and more deeply colored. The head, thorax, antennae, and legs are yellowish or reddish brown, with the upper surface of the head, posterior border of pronotum, three large spots on the mesonotum, disk of scutellum, meso- and metapleurae, and gaster dark brown. Whole surface of body very smooth and shining. Pilosity similar to that of the worker, but longer on the gaster. Pubescence also somewhat longer but not obscuring the shining surface. Wings grayish hyaline, with brown veins and stigma.

MALE. Length 7–9 mm.

Differing from the male of the preceding forms in coloration. The head is black; the mandibles, scapes, thorax, and petiole brownish yellow; the funiculi and gaster dark brown, the genitalia yellow and more or less infuscated. The pleurae, scutellum, and epinotum are spotted with fuscous and there are three large, elongated fuscous spots on the mesonotum. Antennal scapes and tarsi sometimes brown. In some specimens the whole mesonotum is fuscous and in others the lighter portions of the thorax are brown instead of yellow. In still other specimens the whole thorax is dark brown with yellowish sutures. Wings as in the female.

TYPE LOCALITY.— District of Columbia (Th. Pergande).

Virginia: (Th. Pergande).

North Carolina: Black Mountains (Wm. Beutenmüller).

New Jersey: Halifax (Wheeler).

New York: Mosholu (Wheeler).

Pennsylvania: Beatty (P. J. Schmitt).

Connecticut: Colebrook (Wheeler); New Haven (Butrick); Salisbury, New Haven, Orange (W. E. Britton).

Massachusetts: Wellesley (A. P. Morse); Essex County (G. B. King); Forest Hills, Blue Hills, Woods Hole (Wheeler); Arlington (Mus. Comp. Zoöl).

Indiana: Hammond, Kosciusko County, Marion County (W. S. Blatchley).

Illinois: Algonquin (W. A. Nason); Rockford (Wheeler).

Colorado: Manitou, Colorado Springs, Colorado City (Wheeler).

New Mexico: Las Vegas (Wheeler).

Quebec: Hull, near Ottawa (Wheeler).

Ontario: Grimsby (Wheeler).

Emery regards as the type of this subspecies "workers, which have about the color of the subsp. *schaufussi*," but such individuals are too pale to represent the subspecies properly, which is decidedly darker. It is in fact often so dark as to merge into *fuscata*. Such transitional forms are also cited by Emery from South Dakota but he evidently regarded them as *fuscata*. Emery refers the male and female described as *schaufussi* by Mayr to this subspecies.

F. nitidiventris closely resembles *schaufussi* and *incerta* in habits, but nests in more shady situations, along the borders of woods, etc. In geographical range it seems to coincide very closely with *incerta*, from which it is sometimes distinguishable only with difficulty.

138. F. (N.) PALLIDEFULVA NITIDIVENTRIS var. FUSCATA Emery.

F. pallidefulva subsp. *fuscata* Emery, Zool. jahrb. Syst., 1893, **7**, p. 656, ☿ ♀.
F. pallidefulva nitidiventris var. *fuscata* Wheeler, Bull. Amer. mus. nat. hist., 1904, **20**, p. 370.

WORKER. Length 4–6 mm.

Characterized by deeper coloration and feebler pilosity. The body is dark reddish brown or blackish, the anterior portion of the head and legs paler; mandibles, antennae, tarsi, tibiae, and articulations of legs red or yellowish. Tips of funiculi infuscated. The surface of the body is sometimes more sharply shagreened and therefore somewhat more opaque than in *nitidiventris*, the hairs even sparser on the head and usually wanting on the thorax. The pubescence is very short and sparser and much as in *nitidiventris*.

FEMALE (DEÄLATED). Length 7–9 mm.

Dark reddish brown or blackish; mandibles, scapes, pronotum, petiole, legs, and sometimes also the mesonotum yellowish. Hairs more abundant and longer than in the worker, present also on the thoracic dorsum. Surface of body shining, much as in the female of *nitidiventris*.

TYPE LOCALITY.— Pennsylvania: Beatty (P. J. Schmitt).

North Carolina: Black Mountains (Wm. Beutenmüller); Lake Toxaway (Mrs. A. T. Slosson).

Georgia: Thunderbolt, Savannah (J. C. Bradley).

New Jersey: Halifax (Wheeler).

New York: Bronxville (Wheeler).

Massachusetts: Essex County (G. B. King); South Natick (A. P. Morse); Forest Hills, Blue Hills, Woods Hole (Wheeler).

Illinois: Rockford (Wheeler).

South Dakota: Hill City (Th. Pergande).

New Mexico: Las Vegas (Mrs. W. P. Cockerell).

Ontario: Guelph (W. H. Wright).

This form is regarded by Emery as a distinct subspecies, but in my opinion it is hardly more than a melanic variety of *nitidiventris*. Unlike the preceding forms it nests only in woods, usually in hilly country and is much rarer than any of the other varieties or subspecies.

139. F. (N.) MOKI Wheeler.

F. moki Wheeler, Bull. Amer. mus. nat. hist., 1906, **22**, p. 343, ☿.

WORKER. Length 4–5.5 mm.

Mandibles 8-toothed. Maxillary palpi very long, 5-jointed. Head, excluding the mandibles, decidedly longer than broad, narrower in front than behind, with straight posterior border and sides. Eyes large and convex. Clypeus strongly carinate, its anterior border rounded, projecting. Frontal carinae but slightly diverging behind Antennae long and slender; scapes scarcely curved at the base; middle funicular joints more than 1½ times as long as broad. Thorax long and narrow, in profile very low; pro- and mesonotum much depressed, mesoëpinotal constriction shallow and very long at the bottom. Epinotum with straight, horizontal, basal surface, nearly twice as long as the very sloping declivity. Seen from above the pronotum is as long as broad, mesonotum nearly twice as long as broad. Petiole narrow, thick at the base, with sharp horizontal border, and both the anterior and posterior surfaces, but especially the latter, distinctly flattened, so that the segment is cuneate in profile. Gaster small. Legs long and slender.

Opaque, finely shagreened; even the mandibles and frontal area only slightly lustrous; the former finely and densely striated and coarsely punctate. Head behind with a bronzy or glossy surface.

Hairs white, sparse, pointed on the upper surface of the head, obtuse on the gaster; absent on the gula, petiole, and upper surface of the thorax. Legs with only the series of oblique bristles on the flexor surfaces of the tibiae. Pubescence grayish, fine and rather dense, covering the whole surface of the body and appendages, longest on the gaster.

Dull reddish yellow; gaster, posterior half of head above, terminal

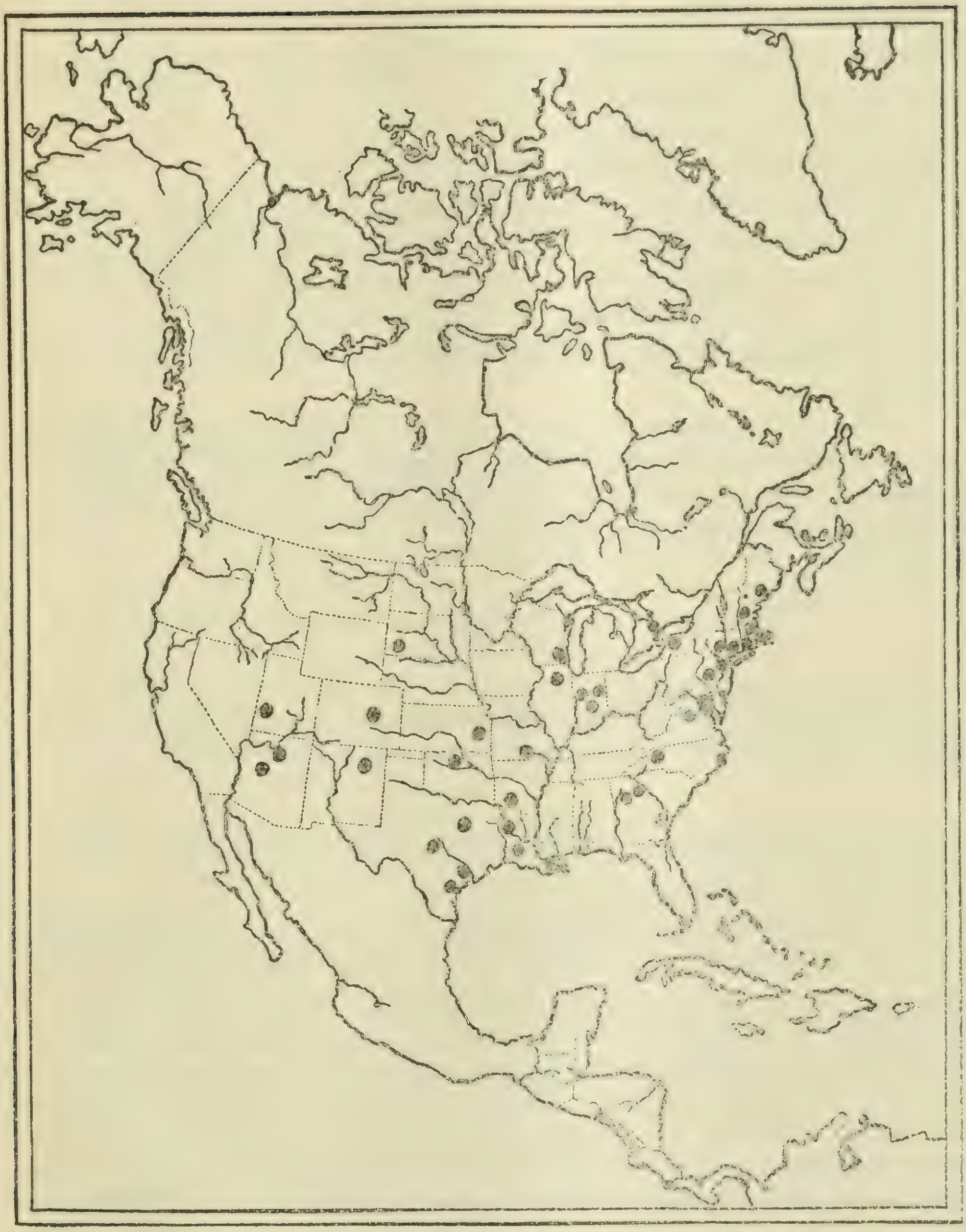

Fig. 10.— Distribution of the subgenus Neoformica.

joints of funiculi, a large spot on the pronotum, a smaller one on the mesonotum, the upper surface of the petiole, the coxae, femora and in some specimens also the apical half of each tibia and the pleurae, dark brown or fuscous.

Type locality.— Arizona: Bright Angel Trail, Grand Canyon 5,500–7,000 ft. (Wheeler).

Arizona: Prescott (Wheeler).

Utah: Milford (J. C. Bradley).

This species appears to belong in the *pallidefulva* group, although the sculpture of the body is very unlike that of the preceding species. Superficially it resembles *F. rufibarbis*, but the head, thorax, and antennae are much more like those of *pallidefulva*. There is, however, much that recalls Myrmecocystus in the structure of the thorax seen in profile.

F. moki nests under stones and forms colonies about the size of those of *F. pallidefulva* and its various subspecies and varieties. It probably represents this species in the dry deserts of the southwest, but is certainly a much rarer ant.

Addendum.

The following notes and descriptions relate to specimens discovered after the manuscript of this paper was ready for the press.

140. F. truncicola integroides var. ravida, var. nov.

Worker. Length 4–6 mm.

Like the var. *haemorrhoidalis* Emery in pubescence and sculpture and in lacking erect hairs, except on the gaster, but differing in color. The red of the head, thorax, and petiole is much deeper and not yellowish, the legs, whole of the funiculi, tips of the scapes and the gaster are black and even the largest workers have a large black spot on the pro- and mesonotum. In small workers the red color is darker and more brownish, the whole thorax is blackish and the posterior portion of the head and the whole of the scapes are infuscated. The surface of the body in all of the workers is opaque, the pubescence on the gaster short, dense and dark gray in color; the red anal spot is much restricted.

Female. Length 8.5–9 mm.

Differing from the female of *haemorrhoidalis* in having the tips of the scapes, the posterior border of the pronotum, three spots on the mesonotum, the whole of the scutellum and metanotum, a few spots on the mesopleurae and the middle and hind legs, including their coxae, black.

Described from two females and seven workers collected by Mr. W. M. Mann at Elkhorn, Montana. He has also taken six workers at Helena in the same state. The two females are immature so that the red of the head and thorax is paler and more yellowish than in the workers. The opacity of the body and the character of the pubescence on the gaster in both female and worker show clearly that this variety is to be referred to the subspecies *integroides* Emery and not to *integra* Nylander.

141. F. SUBPOLITA var. FICTICIA, var. nov.

WORKER. Length 3–6 mm.

Very closely resembling the typical form from California but differing in having the head less deeply and less extensively infuscated behind, the thorax bright red and rarely infuscated even in the small workers, the erect hairs, especially on the pronotum and gula, less numerous, the petiolar border sharper and more compressed, and the antennal scapes a little less enlarged towards their tips.

FEMALE. Length 8.5 mm.

Differing from the female of the typical form in having the clypeus, cheeks, and pleurae red, the mesonotum less shining and the wings somewhat shorter and more nearly colorless.

MALE. Length 7.5–8 mm.

Differing from the male of the typical form in color, the gaster being black, instead of reddish yellow, with the genital appendages more or less infuscated or black. The stipes of the genitalia are broad and blunt, the subgenital plate broad, the gaster compressed dorsoventrally The head is shaped as in the typical form, the wings paler.

Described from one female, five males, and twelve workers taken by Mr. W. M. Mann at Helena, Montana. He has also given me eight workers from Elkhorn in the same state. It is interesting to find this form so far inland from the Pacific Coast. The discovery of the male is somewhat disconcerting, since it would seem to indicate that after all *F. rufiventris* Emery may not be, as I have stated, the male of the typical *subpolita* Mayr, but the male *ficticia* agrees so closely with the form described by Emery, except in the color of the gaster, that I am not ready to admit myself mistaken, especially as the females of the typical *subpolita* vary from black to yellowish red in the color of the gaster.

142. F. MICROGYNA RASILIS var. PULLULA, var. nov.

WORKER. Length 3.5–6 mm.

Differing from the typical *rasilis* in color, the red portions of the body being decidedly darker and more brownish red. The petiole is more compressed anteroposteriorly, with a sharper border, which is more produced upward in the form of a blunt point. The erect, blunt hairs on the upper surface of the head and thorax, especially on the latter, are shorter and even less numerous. In many specimens they are altogether lacking on the front and thorax.

FEMALE. Length 5 mm.

Differing from the female of *rasilis* in color and in the shape of the petiole. Head, thorax, and gaster dark brown or blackish; mandibles, clypeus, cheeks, and gula dark red; antennal scapes, propleurae, epinotum, petiole, and legs somewhat paler, dull red. Head and thorax opaque, gaster slightly glossy, with very short, rather sparse pubescence. Wings grayish hyaline; stigma light brown, veins paler. Petiole as in the worker, but its border even more produced and attenuated in the middle.

Described from numerous workers and three winged females taken from two colonies at Flathead Lake, Montana, by Prof. C. C. Adams. In the coloration of the worker and shape of the petiole this variety resembles *F. adamsi* Wheeler, but is larger and the head and thorax are at most very faintly and diffusely clouded with fuscous and not spotted.

143. FORMICA MICROGYNA RASILIS var. NAHUA, var. nov.

WORKER. Length 4–6 mm.

Differing from the worker of the typical *rasilis* in having the petiole narrower and its margin distinctly blunter, the erect, obtuse hairs on the head and thorax somewhat more numerous and also present on the border of the petiole and on the gula; the sculpture is somewhat sharper, so that the sides of the head are opaque like the remaining surface, and the color of the gaster is darker and more blackish. Even the largest workers have no infuscation on the ocellar triangle and very rarely have faint blotches on the pro- and mesonotum. The tibiae are naked as in the typical *rasilis*.

FEMALE. Length 6 mm.

Differing from the female of *rasilis* in being a little larger and more robust, in having more numerous erect, obtuse hairs on the head, thorax, and gaster, a blunter petiolar border and in color, which is like that of the worker, with red and not brownish yellow, head, thorax, petiole, legs, and antennae. The wings seem to be a little darker than in the typical *rasilis*.

MALE. Length 7 mm.

Differing from the male of the typical *rasilis* only in having somewhat darker wings and a more opaque gaster.

Described from several specimens of all three phases taken by Mr. W. M. Mann at Guerrero Mill (9,000 ft.) and Velasco in Hidalgo, Mexico, from populous colonies nesting under stones banked with vegetable detritus. These colonies were very sporadic, but each contained a large number of the small, winged females. Mr. Mann also found these females (deälated) in two colonies of *F. subcyanea*, sp. nov. (*vide infra*), thus proving that this is the temporary host of *nahua*.

144. FORMICA SUBCYANEA, sp. nov.

WORKER. Length: 4–5.5 mm.

Closely related to *F. fusca*. Head as broad as long, a little narrower in front than behind, with feebly convex sides, rounded posterior corners and straight posterior border. Eyes rather large. Clypeus sharply carinate, with entire, slightly reflected but not produced anterior border. Antennae rather stout, the tips of the scapes a little thicker and the terminal funicular joints a little shorter than in *fusca*. Shape of thorax, petiole, and gaster as in *fusca*, the petiole having a convex anterior and flat posterior surface and a broadly rounded, entire upper border.

Body, including the appendages, opaque, very coarsely shagreened and in this respect resembling *F. fusca* var. *japonica*; gula and mandibles a little more shining, the latter coarsely striatopunctate.

Hairs short, white, erect, a little more abundant on the head and thorax than in *fusca*, obtuse on the gaster. Gula in the middle with a very few erect hairs (1–4). Pubescence yellowish, extremely short, moderately abundant on the head and gaster, less conspicuous on the thorax and appendages.

Deep black throughout, including the antennae, legs, palpi, and mouthparts; only the strigils of the fore tibiae and in some specimens the bases of the scapes, reddish. Body in bright sunlight with distinct, deep metallic blue and bronze reflections.

FEMALE. Length 8–9 mm.

Very much like the female of the typical *F. fusca* in size and shape. Differing from the worker in having longer pubescence on the body and in lacking the metallic blue reflections, though there is more or less of the bronzy effect. The sculpture is as coarse as in the worker or even coarser, especially on the gaster, but the surface of the body, and especially of the scutellum, epinotum, and gaster, is a little more shining. Wings distinctly infuscated, much as in *F. fusca* var. *subsericea,* with blackish veins and stigma.

MALE. Length 9 mm.

Differing from the male of the typical *fusca* in having the wings distinctly infuscated, the bases of the femora and the tips of the external genital appendages more blackish, the surface of the body more coarsely punctate and more opaque and of a deeper black color. The erect hairs on the head and thorax are much more abundant and the pubescence on these parts and on the gaster is distinctly longer and coarser. The mandibles are bluntly dentate.

Described from numerous workers and females and one male taken by Mr. W. M. Mann at Guerrero Mill (9,000 ft.), Velasco, below Real del Monte, El Chico and Pachuca, in Hidalgo, Mexico. Mr. Mann found this ant to be more pugnaceous than *fusca* and its var. *subsericea.* It nests in large colonies under stones in exposed, open localities, such as hill-tops, but more commonly in shady places where the soil is moister.

Were it not for the erect hairs on the gula of the worker and female, and the peculiar sculpture and metallic coloration one would be inclined to regard this ant as a subspecies of *fusca.* It should be placed just after *F. sybilla,* which it resembles in pilosity though it differs in the worker phase in having a stouter body, shorter head, antennae and legs, larger eyes, a much broader petiole and different color and sculpture. The male *sybilla* differs from that of *subcyanea* in having much longer, broader, and more yellowish wings (10 mm. long, as compared with 8 mm. in the latter species), more pilose and more coarsely sculptured head and thorax, broader gaster and somewhat more compressed petiole.

FORMICA RUFIBARBIS Fabr. var. GNAVA Buckley. (Page 518).

Workers and winged females indistinguishable from the more northern specimens of this variety were taken by Mr. Mann at Guerrero Mill and El Chico in Hidalgo, nesting under stones or in mound-nests. The colonies were less populous than those of *F. subcyanea.*

FORMICA CINEREA Mayr var. ALTIPETENS Wheeler. (Page 523).

A few workers found by Mr. Mann running on cactus at Pachuca in Hidalgo agree very closely with the types of this variety from Colorado.

All the foregoing Mexican forms, though occurring as far south as latitude 20° N., were found only at rather high altitudes. In addition to these four forms and *F. perpilosa,* described on p. 421, several other Formicae have been recorded as occurring in Mexico by Forel in the "Biologia Centrali-Americana," namely *F. fusca* var. *subsericea, F. rufibarbis, F. neorufibarbis,* which Forel regarded as a var. of *rufibarbis, F. rufa obscuripes,* and *F. incisa.* Concerning these forms I venture the following remarks:—

1. *F. fusca* var. *subsericea* is recorded from Durango, 8,100 ft. (Forrer), Atoyac in Vera Cruz (Schumann), and Moyoapam (Coll. Saussure). This is much more probably *F. fusca* var. *argentea* Wheeler.

2. *F. rufibarbis* is cited from Sonora (Morrison) and Omilteme in Guerrero (H. H. Smith). This is probably the form which I have called *F. rufibarbis* var. *occidua* and not the typical specific form which is Palaearctic.

3. *F. rufibarbis* var. *neorufibarbis* Forel, which is recorded from Durango, 8,100 ft. (Forrer), is probably the var. *gnava* Buckley.

4. For *F. rufa obscuripes* only the locality "Mexique (Brinkmann)" is given. This form may, perhaps, occur in the high mountains of Northern Mexico, but I am inclined to believe that the specimens to which Forel refers belong to some other *rufa* form or to some member of the *microgyna* group.

5. *F. incisa* was described by F. Smith from a female specimen, with the locality "Mexico." As Forel says, it is an "espèce extrêmement douteuse et indéchiffrable," but it can hardly be the female of some form of *F. rufibarbis,* as he suggests, because the coloration of this species is very different.

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